THE AMERICAN PHYSIOLOGICAL SOCIETY, INC.

FIFTY-SEVENTH ANNUAL MEETING

Atlantic City, New Jersey, March 15, 16, 17, 18, 19, 1948

(For possible corrections in any of the following abstracts see the next issue.)

Cold tolerance and cold immersion in infant rats. E. F. ADOLPH. *Dept. of Physiology, The University of Rochester, Rochester, N. Y.* Rats aged 0 to 17 days recovered after refrigeration, either in oxygen (Fairfield, this issue) or in air, with body temperatures from 3° to 10°C. Such temperatures were tolerated for 2.5 hours, but not for 3 hours. Yet the rats did not tolerate temperatures below 15° when immersed in water to the shoulders. Evidently body temperature was not the only factor in survival of infants. Cold exposure, continued in an atmosphere of nitrogen after the breathing had ceased from cold, allowed no recovery. Surrounded by a rubber jacket held away from the trunk, the infants survived colonic temperatures of 6° while environed by water of 5°. Immersion up to the hips gave similar diminutions of colonic and mouth temperatures, with survival. Such cooling could be followed by immersion to the shoulders for one hour once breathing had ceased, with survival. But gradual cooling of water to 8° while chests were immersed was lethal. Evidently the damage was imposed during the cooling period. Upon rewarming, whether slowly in air or rapidly in water of 35°, the heart regularly resumed beating. Whenever reflexes and breathing also were regained, survival was permanent. Part of the lethal damage may be due to hydrostatic pressure about the chest, and part to local anoxia. Survival of infant rats below 15° depends only in part on their well-known tolerance of anoxia; whereas in hibernants cardiorespiratory functions persist at all temperatures compatible with survival.

Lethality of cold immersion in rats. E. F. ADOLPH. *Dept. of Physiology, The University of Rochester, Rochester, N. Y.* Does failure to survive exposure to cold depend predominantly upon the low temperature to which certain tissues are exposed? Can that temperature be modified? Albino rats were cooled in various ways while their colonic (and sometimes esophageal) temperatures were being measured by thermocouples.

Immersion to the shoulders in water of 14.8°C. for 2 hours was lethal for half of individuals, and none survived in 14.0°. Colonic temperatures at 2 hours were within one Centigrade degree of the water temperature. Reflexes and breathing gradually disappeared; if any remained the rats recovered when placed into air of 25°. Especially when cooling was rapid, reflex and cardiac activities disappeared some time after lethal temperatures were passed, since, in cold, functional breakdowns are delayed. Slow cooling of rats covered by rubber jackets and surrounded by water for 5 hours gave the same lethal temperatures as above. When immersed, uncovered, up to the hips and abdomen, the rats cooled equally slowly, and survived lower colonic temperatures (12° to 14° after 5 hours); but esophageal temperatures were one to three degrees higher. Many individuals died after rewarming with delays of hours or a day; thus they survived only temporarily when colonic temperatures went below 14°C. Median lethal temperatures were not modified by sex, age above 30 days, administrations of glucose or of cortin, deprivations of food or of water, nor repetition of the brief cold exposure. No way is known of securing continued survival of unanesthetized mature rats, after the chest and head have been cooled below 14°.

The reaction caused by the intravenous injection of salmine sulfate in dogs. J. GARROTT ALLEN and WILLADENE EGNER (by invitation). *From the Department of Surgery of the University of Chicago.* Salmine sulfate may be used to control hemorrhage from overdosage of heparin because it forms an inactive salt with heparin. This substance has been said to be toxic, when given intravenously, producing an anaphylactoid reaction. Since this substance agglutinates the red blood cells in certain mammalian species and causes fibrinogen precipitation, the anaphylactoid reaction has been ascribed to the "plugging" of the peripheral vascular bed. Experiments on dogs are presented which show that animals given 5 to 15 mg. of salmine sulfate per kg. of body weight, intravenously, caused an anaphylactoid reaction. Dyspnea and a sharp fall in the arterial blood pressure occurred within two minutes after the injection of salmine. Dyspnea persisted only two or three minutes, but the blood pressure recovered slowly over a period of 30 to 40 minutes. A second injection of the same quantity of salmine sulfate made an hour after the first injection was not followed by any appreciable fall of the arterial blood pressure and dyspnea did not occur. When the animals were given the same amount of salmine in the same manner again one week later, the phenomenon of tachyphylaxis was again observed. Coagulation studies, red cell counts, hematacrit readings, and platelet counts made, showed no significant change after the injections. Leukopenia did occur but the white blood

count returned to normal, as the blood pressure was restored to normal. Following the second injection, when the animal showed no obvious systemic reaction, leukopenia did not occur. At the dosage used, no change in the fibrinogen content was noted after either injection of salmine. Five to ten times the amount employed here was necessary to produce fibrinogen precipitation.

A method for estimating traces of T-1824 by combination with cellophane. THOMAS H. ALLEN and PETER D. ORAHOVATS (by invitation). *Department of Physiology, College of Physicians and Surgeons, Columbia University.* Traces of the blue dye, T-1824, can be estimated quantitatively in the form of its combination with Cellophane. Ninety-eight per cent of the dye present in a 25 ml. solution (0.48% dye diluted 1:10,000 with 0.9% NaCl) is bound on 6.5 mg. of Cellophane foil immersed in the solution for 24 hours at 65°C. The optical density of the dye on the strip is 0.372 at 635 μ. When the concentration is doubled the Cellophane optical density increases 2.08 times. The product of the weight of the Cellophane and the optical density is directly related to the concentration of dye over the dilution range of 0 to 5 parts in 10,000. The homogeneity of staining and the spectral properties of the dye-Cellophane have been studied as a function of time, volume of solution, temperature, pH, concentrations of NaCl and Aerosol OT. This method appears to be selective, reasonably accurate and can be used in urine or blood serum.

T-1824 appears in dog urine within 30 minutes after intravenous injection of the amounts commonly used for blood volume determinations. During the ensuing 24 hours 2 parts in one thousand of the injected dye are excreted by the kidney. This value decreases to 3 parts in ten thousand on the second day. Five drops of dye-tinged dog blood yield Cellophane values which agree well with those obtained by direct spectrophotometric analysis of the serum.

Hemin synthesis with glycine containing C^{14} in its alpha-carbon atom. K. I. ALTMAN, G. W. CASARETT, R. E. MASTERS, T. R. NOONAN, and K. SALOMON, (introduced by W. F. BALE). *University of Rochester School of Medicine and Dentistry, Atomic Energy Project, Rochester 7, New York.* It has been shown by Shemin and Rittenberg (J. Biol. Chem. **155**, 567, (1945); **166**, 621, (1946)) that the nitrogen component of glycine is incorporated in the hemin of hemoglobin. This left unanswered the question whether the carbon skeleton of glycine would also function as a specific precursor of the pyrrol in hemin. In order to investigate the latter point, $C^{14}H_2NH_2COOH$, prepared by B. M. Tolbert (Radiation Laboratory, University of California), was fed to rats which had been rendered anemic by previous phenylhydrazine feeding. These animals were rapidly forming new erythrocytes as indicated by a high reticulocyte count. The radioactive glycine was administered by stomach tube in a single dose of approximately 1 μc/100 gm. body weight. At appropriate intervals animals were bled as completely as possible from the carotid artery. Hemin was prepared following Nencki, M. and Jaleski, J. (Ztschr. f. physiol. Chem. **30**, 384, (1900)), globin according to Anson, M. L. and Mirsky, A. E. (J. Gen. Physiol. **13**, 469, (1930)), and hemoglobin by crystallization after hemolysis. C^{14} was determined according to a method developed by W. F. Bale and R. E. Masters. In a typical experiment the concentration of C^{14} per gram hemin was 9.65 times that found per gram globin. From this evidence we conclude that the alpha-carbon of glycine is a direct precursor of a portion of the hemin molecule.

Extraction of myogen and myosin from whole skeletal muscles. WILLIAM R. AMBERSON, RUBERT S. ANDERSON (by invitation), BETTY CHINN (by invitation) and T. ERDÖS (by invitation). *Department of Physiology, University of Maryland, Baltimore, and Institute of Physical Chemistry, University of Uppsala, Uppsala.* Myogen and myosin may be partially extracted from whole skeletal muscles (rabbit) without mincing the tissues. Whole muscles are placed in closed bottles and quickly frozen in CO_2 snow. After they thaw the extracting solutions are poured over them (1 to 2 cc. per gram). Extraction is rapid in the first day; smaller increments of protein continue to appear for many days. Extraction proceeds best at 0°C. After several days the solutions are poured off, centrifuged, and dialyzed against phosphate buffers. Protein solutions thus prepared show the following characteristics: (1) Distilled water extracts contain myogen up to 2% of the wet weight. In extracts of white muscle the electrophoretic patterns, in phosphate buffer at pH-7.6, show a large, slow component, and a small, fast one. In red muscle extracts the same components occur, plus a myoglobin elevation, intermediate in velocity. These extracts contain no myosin, and have a low viscosity. (2) 0.4M K phosphate buffer at pH-7.6 extracts more protein, but the electrophoretic patterns are still simple, consisting of the myogen components just described. No myosin appears for many days. (3) If $Na_4P_2O_7$ (500 mgm. %) is added to the phosphate buffer, myosin is extracted from whole muscles, both red and white, so that the solutions become very viscous. The pyrophosphate also increases the extraction of the myogen components. Sharp myosin elevations appear in both the electrophoretic and ultracentrifuge sedimentation diagrams. Actomyosin and actin are not extracted. The myosin may be separated from the other proteins by electrophoretic fractionation.

A study of various methods of rewarming men after exposure to extreme cold. ADELBERT AMES III (by invitation), RICHARD S. GRIFFITH (by invitation), DAVID A. GOLDTHWAIT (by invitation), MARTIN B. MACHT (by invitation), and H. S

BELDING. *The Quartermaster Corps, Climatic Research Laboratory, Lawrence, Mass.* To determine the efficiency of various methods of rewarming individuals after exposure to extreme cold, oxygen consumption, skin temperatures, rectal temperatures, thermal comfort and onset of shivering of four men were determined during 15 experiments, each consisting of a one-hour exposure to −40°F., followed by one hour of rewarming and one hour of re-exposure to −40°F. Rewarming was attempted by: removal of subject to higher ambient temperature, i.e., 40°F. or 80°F.; moderate or strenuous exercise, i.e., walking on treadmill 2.5 mph at level or 3.5 mph at 6.5% grade; providing additional thermal insulation (heavy sleeping bag); infrared irradiation of hands or face. The order of efficiency of various rewarming methods, as measured by change in total body heat, was: (1) strenuous exercise, (2) ambient temperature of 90°F., (3) sleeping bag, (4) ambient temperature of 40°F., (5) moderate exercise, (6) irradiation of face, (7) irradiation of hands. The first four procedures caused increases in total body heat during the rewarming period, but only in the first was the original heat content approximated. Moderate exercise resulted in cessation of heat loss without gain during rewarming. Irradiation of hands or face had no significant effect on total heat content. In all cases, body heat decreased during the second hour of cold exposure at approximately the same rate as during the first hour. The only indications of acclimatization, based on results of control experiments, were a progressive increase in thermal comfort and a progressive delay in onset of shivering during successive exposures.

Relation between synaptic delays and characteristics of preganglionic volleys initiating responses in cat autonomic ganglia. DOMINGO AMPIL (by invitation) and HARRY GRUNDFEST. *Dept. of Neurology, College of Pnysicians and Surgeons, Columbia Univ., New York.* Synaptic delays in the superior cervical, stellate, and inferior mesenteric ganglia of the cat have been determined in relation to the type of initiating preganglionic impulse, which may be either of the A or the B fiber type. Both fiber types were found in the preganglionic nerves to the 3 ganglia, but their relative importance varied in different preparations. Six successful preparations of each ganglion, with minimally damaged circulation were studied. Two or all 3 ganglion preparations were made in 4 animals.

Afferent activity was produced by graded electrical stimulation of the preganglionic nerves. Simultaneous oscillographic records of the electrical activity of the preganglionic and postganglionic nerves and of the ganglion permitted correlation of the time relations of the ganglionic response with the conduction velocity of the initiating preganglionic volley. Because of the known relation between fiber velocity and spike duration these experiments afforded a test of the relative importance of the duration of the afferent spike in determining synaptic delay.

Preganglionic volleys beginning with fibers conducting at 34.8 to 17.5 mps., therefore of the A type, with spike durations approximately 0.5 msec., produced ganglionic responses after synaptic delays of 1.9 to 4.1 msec. Preganglionic activity if predominantly B group fibers (volleys with highest velocities of 15.8 to 6.2 msec.; spike durations about 1.2 msec.) produced ganglionic responses after delays of 2.5 to 5.0 msec. Therefore, the nature and duration of the initiating spike apparently are not the primary determinants of synaptic delay in autonomic ganglia.

Changes in nitrogen distribution in the Japanese beetle during metamorphosis. JOHN M. ANDERSON. (introduced by OTTO M. HELFF). *Dept. of Biology, Brown Univ., Providence, R. I.* Lacking histological data, an attempt was made to trace by chemical methods the processes of histolysis and histogenesis during metamorphosis of the Japanese beetle. Analyses of individual insects at all metamorphic stages yielded data for (1) insoluble protein and chitin N, (2) amino-acid N. (3) soluble protein, proteose and peptone N, and (4) uric acid N. These values were expressed in percent of total N at each stage and their fluctuations charted. A conspicuous change occurs at pupation: Fraction (1) decreases, amino-acid N and Fraction (3) suddenly increasing. Changes in the reverse direction have occurred by the 3rd pupal day, and late-prepupal levels are then gradually reconstituted. Uric acid N reaches its peak by the 3rd pupal day and apparently shows no further changes. No differences were found between late pupae and adults.

It is assumed that decrease in insoluble N and increase in soluble N indicates histolysis, reverse changes indicating histogenesis. The reciprocal shifts in these fractions at pupation do not accompany, but follow, the fall in oxygen consumption and in blood pH found by Ludwig (1931, 1934). These latter changes may be considered as manifestations of processes occurring prior to disintegration of larval tissues at pupation.

Chloruretic action of pressor-antidiuretic fraction of posterior pituitary extract. W. PARKER ANSLOW, JR., LAURENCE G. WESSON, JR., ALFRED A. BOLOMEY and JOHN G. TAYLOR (introduced by HOMER W. SMITH). *New York University College of Medicine.* The chloruretic action of the pressor-antidiuretic fraction of posterior pituitary extract (Pitressin) has been examined under conditions which preclude contribution to chloruresis by the oxytocic principle (Pitocin) and by changes in filtration rate and plasma sodium and chloride. In 5 of 6 experiments on 2 dogs under water load (presumaably minimal endogenous posterior pituitary secretion) and under conditions otherwise conducive to

stable chloride excretion, the rate of excretion on discontinuing Pitressin infusion fell to a level one-half or less the maximal rate during Pitressin administration (15–20 mμ. per hour). Changes in sodium and potassium excretion generally paralleled changes in chloride excretion. Ammonium excretion was unchanged. In experiments with no water load (presumably maximal endogenous posterior pituitary secretion), no significant change in chloruresis was observed when Pitressin was administered or discontinued. In two experiments the action of Pitressin was compared with that of Pitocin at a dose of the latter which gave little or no antidiuretic effect (26 mμ. of Pitocin per hour). No chloruretic effect was observed. The results indicate that the pressor-antidiuretic fraction is chloruretic, but that this chloruretic action is demonstrable experimentally only when endogenous secretion is minimal (i.e., during water load). No new evidence is adduced as to whether the chloruretic action is attributable to the antidiuretic hormone or not.

Physiological significance of preserved central vision in lesions of the optic tract vs. optic radiation. GEORGE M. AUSTIN (by invitation), F. H. LEWEY and FRANCIS C. GRANT (by invitation). *Lab. of the Neurosurg. Service, Hosp. of the Univ. of Pennsylvania.* The present theory of cortical representation of the macula postulates: macular sparing in the visual field with lesions of the occipital lobe, macular splitting with lesions of the optic tract. It is also believed that a lesion large enough to involve the entire optic radiation or area 17 (striata), will produce hemianopia with macular splitting. Contrariwise, clinical evidence has suggested a bilateral macular representation in the occipital lobes. We have examined persons with complete occipital lobectomy including the entire area striata. Central visual fields were mapped out with the Bjerrum tangent screen and the Lloyd stereocampimeter. These patients showed only a small area of preserved central vision varying from ¼ to 1½°. Central field studies of patients with optic tract lesions revealed similar small areas of preserved central vision. The macula includes an arc of approximately 10°C, the fovea an arc of approximately 2°. Our observations failed to show macular aplitting in optic tract lesions or macular sparing following interruption of the optic radiation, or removal of the striate cortex. The absence of foveal sparing vindicates against a bilateral macular cortical representation, for this pre-supposes the preservation of at least an arc of the fovea. The absence of hemianopia with macular splitting after complete removal of area 17 or the optic radiation may be explained by the use of a slightly eccentric foveal point for fixation.

The role of the bulbar facilitatory and inhibitory systems in vasomotor and respiratory activity. L. M. N. BACH (introduced by H. S. MAYERSON). *Department of Physiology, Tulane University School of Medicine, New Orleans.* The reticular matter of the brainstem has been shown to play a most important part in facilitation and inhibition of reflexly and cortically induced somatic motor activity. Comparison of points in the reticular system producing these effects with those found controlling vasomotor and respiratory activity shows a high degree of coincidence. Simultaneous records were made of the knee-jerk, blood pressure and repiration in the cat while selected points in the reticular matter of the pons and medulla were stimulated. It was found that stimulation of points which caused complete inhibition of knee-jerk always caused a marked drop in blood pressure as well as a total inhibition of all respiratory movements usually persisting for the duration of the stimulus. Following cessation of the inhibitory stimulus, whenever there was a rebound effect on the knee-jerk (post-inhibitory facilitation) there also occurred simultaneously a marked increase in blood pressure and in both rate and depth of respiration. These effects occurred simultaneously for equal periods of time, declining similarly to the normal values. Converse vasomotor and respiratory effects were found in knee-jerk facilitation with post-facilitatory inhibition.

Apparently bulbar facilitatory and inhibitory systems apply equally to vasomotor, respiratory, and somatic reflexes. Since it was impossible to obtain pure vasomotor or respiratory effects without definitely affecting knee-jerk activity, it may be assumed that no completely specific brainstem vasomotor or respiratory centers exist. To varying degrees the reticular matter in these areas seem simply to facilitate or inhibit spinal motor cells in general.

Peripheral vascular effects produced by localized warming of various skin areas. MORTIMER E. BADER, MARTIN B. MACHT, and ELIZABETH L. PILLION (introduced by H. S. BELDING). *The Quartermaster Corps, Climatic Research Laboratory Lawrence, Massachusetts.* By means of a venous occlusion plethysmograph and thermocouples placed on selected skin areas, the effect on blood flow through the hand produced by warming localized body areas was studied. A 250 watt infrared bulb located at the base of an insulated metal cylinder (diam. 20 cm.) and controlled through a rheostat was used as a heat source. The subject was seated comfortably with his left hand in an air plethysmograph at heart level and the open end of the cylinder was applied to the area being warmed, a diaphragm being used to prevent heat leakage. Rectal temperature, hand skin temperature, and skin temperature at five widely separated points were determined during each experiment. Subjects remained inactive at an environmental temperature of 15°C. until hand

skin temperature reached equilibrium (approximately 17°C.), at which time basal blood flows and skin temperatures were measured.

Using two subjects in repeated experiments it was demonstrated that heating the face to 44–45°C. resulted in significant increases in hand skin temperature (average rise 10.2 C.°) and average maximum flows 4 times as great as the basal values. Increases generally became apparent within one hour and continued until termination of the experiment at 90 minutes. No changes in rectal temperature, or skin temperature elsewhere were observed. Control experiments, applying heat to the chest in identical fashion, produced no significant changes in hand skin temperature or blood flow. These results suggest that warming certain skin areas is more effective than warming others in overcoming the vasoconstriction of the extremities produced by cold.

Hepato-renal factors in circulatory homeostasis. XX. Antidiuretic action of liver VDM concentrates. S. BAEZ (by invitation), A. MAZUR and EPHRAIM SHORR. *Dept. of Medicine, Cornell University Medical College and The New York Hospital, New York City.* Extracts of anaerobic liver chemically fractionated to concentrate their VDM component were found to produce a profound antidiuretic effect upon intravenous injection into unanesthetized dogs. The VDM content of the liver concentrates, as assayed by the rat mesoappendix technique, was such that 0.001 to 0.0001 gamma N produced a well-defined vaso-depressor effect in the test rat. The amount of VDM infused into the dog for antidiuretic study varied from about 50 to 200 gamma N per minute, depending on the potency of the particular preparation. The rate of injection was regulated so as to maintain a relatively uniform blood level of VDM sufficient to give a transferrable vaso-depressor effect in the test rat. The infusion was continued for 60 to 90 minutes, a total of 2.0–20.0 mg. VDM N being administered in the different experiments. The amounts injected were for beef liver VDM 7.0–12.0 mg. N, for horse liver 2.5–3.0 mg. N, and for dog liver preparations 2.0–2.5 mg. of VDM N. In one series of experiments a study was made of the effect of VDM in the diuresis produced by the infusion of 5% dextrose. In a second series the effects on water diuresis (500 cc. by stomach tube), were determined. The infusion of the VDM preparations resulted, within 10 to 15 minutes, in a 70–80% reduction in urine flow which was sustained throughout the period of infusion. The relation of the antidiuretic effect to the VDM content of the preparations and to possible extraneous materials will be discussed.

A Geiger-Müller counter system for tracer studies of gas exchanges in man. A. C. BARGER (by invitation), G. S. RICHARDSON (by invitation) and E. M. LANDIS. *From the Department of Physiology, Harvard Medical School.* An atmospheric pressure Geiger-Müller counter (Brown, Good and Evans) has been modified for the purpose of determining the kinetics of gas exchange in lower animals and man. The counter, consisting of an oxidized copper tube and tungsten center wire, has been inserted directly into a closed respiratory circuit. This permits rapid recording of saturation or desaturation with minute amounts of a wide variety of radioactive gases, including A^{37}, the radiation from which is so weak that it is detectable only when introduced into the sensitive volume of the counter itself. An automatic feed-back compensator is incorporated into the system to correct for limited changes in gas composition. A mixture of 80% helium with 20% oxygen has proved to be a much more suitable filling gas than air or oxygen because it requires lower voltage and yields lower background with better reproducibility.

Curves showing the absorption of A^{37} in man may be roughly divided, for preliminary analysis, into three parts:—(a) a rapid decline in counting rate lasting from 1–2 minutes, representing chiefly pulmonary mixing, (b) a much less rapid decline lasting from 4–7 minutes during which argon enters the blood and presumably the more richly vascular tissues, (c) a very slow decline representing more gradual penetration of other regions of the body.

Determination of protein-bound iodine in biological material. S. B. BARKER and H. J. LIPNER (by invitation). *Department of Physiology, State University of Iowa, Iowa City.* A procedure has been evolved, by combining and modifying previous methods, for the determination of protein-bound iodine in various tissues, including blood plasma. The principal steps are: precipitation, washing and oxidation of the protein, distillation of the iodine into sulfite, elimination of SO_2 following acidification, and colorimetric determination of the iodide by means of its catalytic effect on the reduction of ceric ions by arsenious acid. Advantage is taken in the last step of the chloride enhancement of iodide catalysis pointed out by Sandell and Kolthoff. This method permits a satisfactory analysis to be performed on 2 ml. of plasma, one-fiftieth of a normal rat thyroid gland, or 500 mg. of rat liver, kidney, heart or skeletal muscle. The changes in protein-bound iodine produced by injection of thyroxin and of elemental iodine are being studied by means of this procedure.

Inulin, urea, mannitol, and PAH clearance ratios in premature infants. HENRY L. BARNETT (by invitation), HELEN MCNAMARA (by invitation), RUTH S. HARE (by invitation) and KENDRICK HARE. *New York Hospital and the Department of Pediatrics, Cornell University Medical College, New York City.* Simultaneous measurements of 98 inulin and urea clearances and 69 inulin, urea, and mannitol clearances were made in 10 permature infants ranging in weight from 2156 to 2470 Gm.

(surface area from .164 to .182 sq. M.). The inulin U/P ratios ranged from 3.9 to 127 and exhibited a close inverse relationship to the urea/inulin clearance ratios which ranged from 0.25 to 0.95. The average mannitol/inulin clearance ratio was 0.90 ± 0.07. In 8 of the premature infants whose ages ranged from 9 to 21 days, the average inulin and mannitol clearances were 4.37 ± 0.88 and 3.93 ± 0.60 ml./min., respectively. In contrast to the findings of Berger, et al. in adult subjects (Proc. Soc. Exper. Biol. & Med. **66:** 62, 1947), the inulin clearances were not lower after mannitol in 5 infants whose control inulin clearances were measured prior to the infusion of mannitol. PAH clearances with plasma levels less than 2.0 mg./100 ml. and PAH tubular maxima with levels greater than 50 mg./100 ml. were measured in 4 of the above infants who were less than 14 days old. The average PAH clearances and tubular maxima were 13.2 ml./min. and 0.97 mg./min., respectively. If PAH clearances at plasma levels below 2.0 mg./100 ml. measure renal plasma flow in these infants, the average filtration fraction calculated as the inulin/PAH ratio is 0.34. Higher values for all of these measurements in infants of the same weight range (2 to 2.5 Kg.) but over 50 days of age suggest that development of kidney function correlates more closely with postnatal than with gestational age.

CO_2 content of maternal and fetal sheep blood. DONALD H. BARRON. *Laby. of Physiology, Yale Univ. School of Medicine, New Haven, Conn.* Observations of the CO_2 content of blood in the uterine and umbilical vessels in pregnant sheep in the last half of the gestation period indicate: 1) that the content of the fetal blood is higher than that of the maternal; 2) that the content of the fetal blood tends to increase regularly as gestation advances, whereas 3) no such uniform change has been observed in the CO_2 content of the maternal blood; 4) the CO_2 content of the blood in the umbilical artery tends to be uniformly higher by four or five volumes per cent than blood in the umbilical vein.

Molecular structure and activity of "vitamin P"-like substances. Inhibition of succinoxidase. GRANT R. BARTLETT (introduced by WILLIAM G. CLARK). *Scripps Metabolic Clinic, La Jolla, Calif.*

Elsewhere (Clark and Geissman, Fed. Proc., 1948) it was pointed out that "vitamin P"-like compounds with an ortho-dihydroxyphenolic structure, a side chain containing an unsaturation and a carbonyl, are epinephrine potentiators (antioxidants?).

In considering possible tissue reactions of these substances, it is recalled that the ortho-diphenols are readily oxidized to quinones which in turn could be toxic to enzyme systems requiring —SH groups for their activity. The compound 2′,3,4-trihydroxychalcone was found by Clark and Geissman to be one of the most active epinephrine antioxidants. It gave a strong inhibition of the —SH enzyme succinoxidase, the inhibition increasing at pH values above 7. At pH 7.4, 10^{-4} M chalcone almost completely inhibited the enzyme. The cytochrome-C-cytochrome oxidase component of the enzyme system was not affected. Glutathione prevented but did not reverse the inhibition. When one of the ortho-dihydroxy groups is covered as in 4-methoxy,2′,3-dihydroxy-chalcone, a quinoid form could not be obtained on oxidation, and succinoxidase is not affected. In the light of the known reactions of quinones, the evidence favours the view that the enzyme inhibition is due to a quinoid oxidation product.

Relationship between arterial pressure and renal blood flow. WOODROW BATTEN by invitation), BEN C. OGLE (by invitation), CARLOS RAPELA (by invitation—Rockefeller Fellow), J. ROY HEGE, JR. (by invitation), J. MAXWELL LITTLE, HAROLD D. GREEN. *Department of Physiology and Pharmacology, Bowman Gray School of Medicine of Wake Forest College, Winston-Salem, N. C.* A kidney and the distal stumps of the aorta and vena cava were progressively dissected free from a dog (kidney dog) until they were connected only by the proximal portion of the aorta and vena cava. A donor dog was heparinized and its femoral artery connected with the distal stump of the kidney dog's aorta. The distal stump of the kidney dog's vena cava was connected with an outflow meter. The proximal stumps or the kidney dog's aorta and vena cava were then ligated and severed thus completing the isolation of the kidney without its being at any time subjected to ischemia. The perfusion pressure was controlled by an air-chamber and automatic electric clamp on the femoral artery of the donor dog. The pressure was lowered for periods of one-half to fifteen minutes to a given level and then returned to the control pressure. Control flows averaged 230 ml./min./100 gm. of kidney (range 115 to 450) at arterio-venous differences of pressure of 120 mm. Hg. Satisfactory plots of flow vs. A-V difference of pressure were obtained in nine experiments. Of these only one gave a curve concave to the pressure axis as described by Selkurt. Two were convex and the remainder were straight. All tended to intercept zero flow at approximately 20 mm. Hg pressure. In this isolated kidney preparation no evidence of autoregulation of flow was observed.

Preliminary studies suggest that a permanent decrease in PAH clearances occurred early in the course of these experiments. The cause of this is being investigated.

A further study of the effect of hemoglobin on cellular respiration. J. PERCY BAUMBERGER, GEORGE F. LEONG (by invitation) and KATHLEEN BARDWELL (by invitation). *Physiology Depar*

ment, School of Medicine, Stanford University. Yeast cells increase their oxygen consumption rate at low oxygen tensions when oxyhemoglobin and hemoglobin are added to the suspension. Several working hypotheses were tested in seeking an explanation of this phenomenon which has been studied by Neumann (1935), Emodi and Sarkamy (1937) and Baumberger (1938). The hypotheses may be stated as follows:—*The increase in Q_{O_2} of yeast on the addition of oxyhemoglobin-hemoglobin is due to:*—(1) an improved oxygen supply to the yeast cells because of the liberation of oxygen from the oxyhemoglobin, or (2) the catalytic action of the methemoglobin that may be formed, or (3) the liberation of some component of the erythrocyte, other than hemoglobin, or (4) the hemoglobin itself. Spectrophotometric and polarographic methods were employed in testing out these hypotheses. The first three are untenable but the fourth hypothesis has merit. So far it can be said that oxy hemoglobin-hemoglobin increases the Q_{O_2} of yeast whether it is in the intact erythrocyte or freed by hemolysis or is purified but it has no effect if oxidixed to methemoglobin or combined with carbon monoxide. The percentage increase in Q_{O_2} is a linear function of the log of the percentage hemoglobin in the oxyhemoglobin-hemoglobin mixture.

The femoral A-V glucose differences following glucose infusion in unanesthetized normal and adrenalectomized dogs. C. H. BEATTY (Introduced by MAGNUS I. GREGERSEN). *Department of Physiology College of Physicians and Surgeons, Columbia University.* It is generally agreed that the disappearance rate of injected glucose in adrenalectomized animals in good condition is at least as rapid as in normal animals. However, it is not clear whether the sites of disappearance are identical. In the following experiments the femoral A-V glucose differences were measured in 6 control and 4 bilaterally adrenalectomized dogs in good condition. Glucose (0.75 gm./kgm.) was injected intravenously in 10 cc. of water and blood samples were taken at 15, 30, 45 and 60 minutes. A statistical analysis of the femoral A-V glucose differences following glucose infusion showed that these were the same in normal and adrenalectomized animals. The fact that no significant variations in blood flow were found after glucose injection in 2 heparinized control dogs (± 10% by bleed out method) indicates that glucose itself produces no changes in the rate of blood flow. But since no blood flow measurements were made upon the adrenalectomized dogs this evidence on the rate of glucose utilization is incomplete.

Oxyhemograph studies during marked vasomotor changes. VIVIAN G. BEHRMANN (introduced by ROBERT GESELL). *Department of Laboratories, Henry Ford Hospital, Detroit, Michigan.* The photoelectric oxyhemograph recently described by Hartman, Behrmann, and Chapman is so constructed that vasomotor changes are cancelled out in the continuous recording of O_2 saturation under ordinary circumstances. No evidence of vasomotor disturbances has been observed in a series of anesthesia studies on experimental and clinical subjects. However, fluctuations in the O_2 satuartion curve have been induced in the experimental animal on the administration of vasodilator (histamine) and vasoconstrictor (adrenalin) drugs, in amounts sufficient to create marked blood pressure alterations. This prompted a search for the factors responsible for these variations. Blood volume changes at the site of the photocell as well as analyses of the blood draining this area should offer valuable information in the solution of this problem. Therefore, all veins, except the main trunk of the anterior auricular vein, were tied off at the base of the ear a day or two previous to the experiment. Blood volume flow was registered from the cannulated anterior auricular vein through a Gibbs drop recorder. The blood, draining the ear photocell site, was collected under oil and analyzed for comparison with the O_2 saturation values obtained on arterial blood by Van Slyke's manometric method. The effects of various vasoconstrictor and vasodilator agents on the blood oxygen saturation curve, coincident with tracings of mean blood pressure, pulmonary ventilation, and blood volume flow from the photocell site are shown.

The tolerance of dogs to intravenously administered fatty chyle and synthetic fat emulsion. ISAAC MURPHY BERRY (by invitation) and A. C. IVY. *Dept. of Clinical Science, University of Illinois College of Medicine, Chicago.* Doses of one gram of fatty acids per kilogram of body weight were given 10 fasting dogs intravenously over a period of about 12 minutes in the form of fatty chyle collected from cannulated thoracic ducts of donor dogs, and in the form of the 10% butter oil emulsion prepared according to the method of Freeman and Meng (average particle size less than one micron). The average level of blood fatty acids for the 10 dogs, expressed as milligrams percent of oleic acid at 0, 0.5, 1, 1.5, 2.5, 3.5, 4.5, 5.5, 6.5, 7.5, and 8.5 hours after injection of the synthetic fat emulsion was 204, 450, 437, 410, 365, 362, 324, 295, 295, 277, and 232 compared to 212, 537, 365, 313, 212, 202, 206, 211, 217, and 186 after the injection of the same amount of fat as chyle. The disappearance from the blood stream of the injected fat was significantly more rapid when the fat was in the form of chyle than when it was in the form of the synthetic emulsion.

Cerebral cortical effects of curare. RICHARD G. BERRY, LIEUT. (MC) US Navy, and FRANCIS M. FORSTER (Introduced by M. H. F. Friedman.) *Dept. of Neurology, Jefferson Medical College, Philadelphia.* The application of curare to the ex-

B & Z Library

posed cerebral cortex of cats, previously anesthetized with Dial, results in the appearance of increased electrical activity of the cortex within four to six minutes. This increased activity is manifested in the electrocorticogram by diphasic spikes similar to those obtained with strychnine, acetylcholine, metrazol and picrotoxin. When the curare is applied to area 7, these spikes show typical transneuronal firing to the contralateral area 7, and when applied to the acoustic cortex it is possible to "drive" the spikes by appropriate stimuli in an anologous manner to the acoustic driving of strychnine and acetylcholine discharges. There is no preliminary cortical depression as noted with ACh. When applied to the motor cortex only minimal and inconstant skeletal muscle response is noted. Varying the pH of the solution did not alter the discharges produced by Intocostrin (Squibb) and d-tubocurarine chloride. It is concluded that curare, applied directly to the cerebral cortex of cats, has a stimulating effect on the electrical activity of the cortex.

A new concept of phase-boundary potential applied to the electro-physiology of nerve. R. BEUTNER and T. C. BARNES. *Dept. of Pharmacology, Hahnemann Medical College and Hospital of Philadelphia.* The rapidity of the rise of electrical potential produced by an alkaloid added to the phase boundary between an oil layer and an aqueous medium suggests that the spike potential in living nerve may be a phase boundary potential (Barnes and Beutner, Science **104:** 569, 1946; Barnes, Fed. Proc. **6:** 73, 1947; Seventeenth International Physiol. Congress 390, 1947). By the courtesy of Dr. H. K. Hartline cathode ray oscillograph records were obtained with a DC amplifier used for electroretinograms. The addition of 0.07 cc. of 0.0027 M acetylcholine chloride in saline to a guaiacol interface in 200 cc. of saline produced a wave of negativity of 2 millivolts lasting 0.2 of a second. This wave of negativity which resembles the action current in living nerve was followed by small positive and negative components not unlike after-potentials. Like the spike potential in nerve the acetylcholine potential is reversible as indicated by the return of the wave to the base line and by the following conductivity measurements. 20 cc. of guaiacol was shaken for two hours with 100 cc. of 0.0027 M acetylcholine chloride in saline and the electrical resistance of the oil was 5.75×10^5 ohms compared with $> 2.5 \times 10^6$ ohms for guaiacol control shaken with saline. The oil containing the acetylcholine was then reshaken with saline and the resistance returned to the original value of $> 2.5 \times 10^6$ ohms. These investigations afford additional evidence disproving the old membrane theory of bioelectrical potential in nerve.

Survival of adrenalectomized-nephrectomized rats treated with desoxycorticosterone. JAMES H. BIRNIE (by invitation), W. J. EVERSOLE (by invitation) and ROBERT GAUNT. *Department of Zoology, Syracuse University, Syracuse, N. Y.* Studies on the extra-renal action of desoxycorticosterone acetate (DCA) in water intoxication (in press) led to observations on the effect of this steroid on the survival of nephrectomized animals; also marked differences in the survival of rats of different ages after nephrectomy were observed. In male rats weighing approximately 130 grams, the right adrenal and kidney were removed in one operation and the left adrenal and kidney removed one week later. One group was given 3 mgm. of DCA per day beginning immediately after the second operation. A second group received 0.5 cc. of peanut oil in a similar fashion. In a third group the kidneys but not the adrenals were removed. No treatment was given any group prior to nephrectomy.

Adrenalectomized-nephrectomized rats treated with DCA survived on the average 51.1 hours (9 animals). Adrenalectomized-nephrectomized rats given peanut oil survived 30.3 hours (10 animals). Rats which had been nephrectomized only survived 29 hours (20 animals). These results indicate that, for the size of rat used, DCA exerts a protective action against the effects of nephrectomy which is greater than that offered by the intact adrenal gland.

In rats that were nephrectomized and untreated those weighing 81 to 120 grams survived 27 hours (12 animals), those between 240 and 322 grams survived 64 hours (8 animals), and those in between these extremes showed an intermediate survival time (11 animals). These results explain some of the wide discrepancies in the literature concerning survival times of nephrectomized rats.

The recovery of sodium thiocyanate from whole blood and plasma as related to the measurement of extracellular fluid volumes. E. W. BIXBY (introduced by H. C. BAZETT). *Physiology Department, Medical School, University of Pennsylvania, Philadelphia, Pennsylvania.* It has been found that extracellular fluid volumes, using the sodium thiocyanate method, increased in malarious humans with the severity of the infection (Overman, Davis, and Thorpe, Amer. Physiol. Soc. Fed. Proc. **6:** 1, pt 2, 174, 1947). In order to study the permeability of the malarious erythrocyte to thiocyanate, normal whole blood has been used as a preliminary control. Whole blood and plasma were studied separately in their reactions to both concentrated and dilute solutions of sodium thiocyanate. When the concentrated solution of thiocyanate (4.71%) was mixed with blood and plasma, the final concentration of thiocyanate was 13.5 mgm./cc. With plasma all the thiocyanate was recoverable within the limits of accuracy (1–2%). With whole blood equilibrium between erythrocytes and thiocyanate

was found when the thiocyanate concentrations of the separated erythrocytes and plasma were analyzed separately. Dilute thiocyanate .01% gave a resultant concentration of .05 to .025 mgm. per cc. in plasma and blood. These concentrations approach the usual range of thiocyanate level found in the serum of subjects on whom extracellular fluid volumes are determined by the thiocyanate method (Crandall and Anderson, Am. J. Digest. Dis. & Nutrition, **1**: 126, 1935).

When plasma alone was used, 10–20% of the thiocyanate added could not be recovered. In these dilute concentrations, small increases of thiocyanate added to a constant plasma volume gave higher percentage values for thiocyanate recovery. And the reverse was likewise true. When whole blood was used with this dilute concentration of thiocyanate only 5–10% of it could not be recovered. If the loss of thiocyanate due to plasma alone is considered, this is evidence that equilibrium between erythrocytes and thiocyanate is established in dilute concentrations.

A light-stable visual purple. Alfred F. Bliss. *Department of Physiology, Tufts College Medical School, Boston.* Squid retinas contain cephalopsin (Biol. Bull. **91**: 220), a visual purple similar to that of vertebrates in its absorption spectrum and in that it can be bleached thermally or by polar solvents, yielding a lipid called indicator yellow, which in acid solution releases the petroleum ether-soluble carotenoid, retinene. However, when dark adapted squid retinas are illuminated continuously one hour after excision into petroleum ether, they show neither bleaching nor increased retinene release in light intense enough to bleach a vertebrate retina in less than a minute. On the other hand, when squid retinas are illuminated immediately after excision into petroleum ether, the thermal rate of retinene release is doubled and maintained for more than three hours. It is concluded, from the maintenance of the light-initiated retinene release in spite of the demonstrated absence of a light-sensitive retinene precursor in one hour old retinas, that there is no present evidence for a photolabile visual purple in the squid. It is suggested that the light effect is an acceleration of thermal bleaching caused by the accumulation of polar metabolic products in the stimulated retina.

The source of uropepsin in man. Stanley Block (by invitation), Louis Rosenberg (by invitation), R. H. Broh-Kahn (by invitation) and I. Arthur Mirsky. *May Institute for Medical Research of the Jewish Hospital, Cincinnati, Ohio.* The urine of subjects with an intact, functioning stomach contains a pepsin-like enzyme which has been designated as uropepsin. An attempt has been made to determine the site of formation of this enzyme and the mode of its entrance into the urine. Previous studies have indicated that this enzyme originates from the stomach. This has been inferred from its absence from the urine of gastrectomized animals and of human patients with pernicious anemia. We have extended these observations by demonstrating its absence from the urine of gastrectomized patients. Although uropepsin originates from the stomach it appears to be absorbed directly into the blood stream from the secreting peptic glands rather than after its excretion into the lumen of the stomach. This was demonstrated by feeding large amounts of gastric pepsin to patients with pernicious anemia, to normal human subjects and to dogs. In no case did this procedure result in an elevation of the levels of uropepsin excretion which had existed prior to the oral administration of the enzyme. The intravenous administration of pepsin to dogs did not result in an increase of uropepsin excretion. Such an observation indicates the inability of pepsin to be excreted after its introduction into the blood and is presumptive evidence that the enzyme is transported intravascularly in the form of pepsinogen, a finding which also would tend to confirm the hypothesis that the enzyme is absorbed directly from the secreting cell in the form of its inactive precursor, pepsinogen.

A method for production of cardiac infarction in the dog. J. Richard R. Bobb, Donald C. Kunze and Wm. McCall, Jr. (Introduced by Harold D. Green). *Department of Physiology and Pharmacology, Bowman Gray School of Medicine of Wake Forest College, Winston-Salem, North Carolina.* Simple ligation of a coronary artery or a coronary artery and its accompanying vein has given irregular unpredictable areas of necrotic myocardium and at times no necrotic myocardium could be found at all. Since the major vessels of the heart are on the surface it was felt that possibly the collateral circulation which supplies the myocardium after ligation of its primary supplying artery has its main branches, although very small, also on the surface. To test this idea dogs were anesthetized with intravenous sodium pentobarbital (40 mg./kg.); artificial respiration was administered and aseptic thoracotomies performed. Ligation of the anterior descending arteries and accompanying veins was done in ten dogs. The arborizations of the ligated vessels were exposed and a cautery touched lightly around the area supplied by the ligated artery. The chests were closed aseptically. All dogs showed definite gross infarcts when sacrificed about ten days postoperatively. In eight control animals the anterior descending arteries and veins were ligated similarly to the test animals. No cauterization was done. Two of these showed extensive infarcts. Four showed infarctions of varying size and two showed no evidence of infarction. Four controls were done in which the only cardiac lesion was a superficial burn around the myocardium supplied

by the anterior descending artery. None of these animals showed infarction. For three days postoperatively a total of 100,000 units of penicillin was administered in divided doses. In a series of 25 dogs with thoracotomies there were no deaths from infection.

It is believed this method will produce a cardiac infarction with greater consistency and of more predictable size than simple ligation.

Change in riboflavin during embryonic development. J. H. BODINE and L. R. FITZGERALD (by invitation). *Zoological Laboratory, State University of Iowa.* A quantitative study has been made of the rate at which riboflavin is converted into pterines during the course of embryonic development of the grasshopper. By the use of x-irradiation it has been possible to destroy parts of the egg and to determine the extent to which embyronic versus non-embryonic cells are responsible for such chemical conversions. In general it has been found that the non-embryonic cells (serosa, etc.) do not change riboflavin into pterines. Such conversions appear to be properties of the embryonic cell *per se*. A chronological table of these conversions has been worked out.

Renal fraction in normal subjects and in subjects with essential hypertension. ALFRED A. BOLOMEY, ERNEST F. BREED, ALEXANDER MICHIE, KATHARINE MICHIE and HENRY D. LAWSON (introduced by H. W. SMITH). *New York University College of Medicine.* Simultaneous determinations of cardiac output (C.O.) by right heart catheterization and renal blood flow (R.B.F.) by the p-aminohippurate clearance method were made on 18 non-hypertensive and 18 hypertensive subjects. The control renal fraction (RBF/CO × 100) averages 15.7 per cent (10.2–26.9 per cent), a value lower than the 19–20 per cent obtained when the average normal resting cardiac output reported by Cournand or by Stead is divided into the average normal value for renal blood flow reported by Smith. This discrepancy may be due to differences in the hematocrit or to hypermetabolism induced by the complexities of the procedures. C.O. showed no correlation with anemia but was increased in realtion to the degree of hypermetabolism. R.B.F. showed no correlation with hypermetabolism but was decreased by anemia. Deviations in metabolic rate and in hematocrit in the hypertensive subjects were of the same order of magnitude as in the controls. The increase in C.O. associated with hypermetabolism and the decrease in R.B.F. associated with anemia operate to lower the renal fraction in both groups. The average C.O. is the same innormal and hypertensive subjects, whereas the R.B.F. varies from normal to very low values in severe hypertension, the renal fraction decreasing to as low as 1.4 per cent in the latter (range 1.4 to 20.7, avg. 11 per cent).

Quantitative glycogen determinations on specimens of human livers obtained by needle biopsy. PHILIP K. BONDY, WALTER H. SHELDON and LILLIAN EVANS (introduced by JAMES V. WARREN). *Departments of Medicine and Pathology, Emory University School of Medicine.* Studies of the liver glycogen concentration in animals have produced valuable information concerning the metabolism of carbohydrates. Comparable studies in human beings have not been carried out, except under distinctly unphysiologic circumstances. Single or serial specimens of human liver tissue can now be obtained by needle biopsy under relatively atraumatic conditions. The histochemical demonstration of glycogen has been improved by Gomori's new method. A quantitative estimation of the liver glycogen content has been made by comparing the optical density of standard sections stained under standard conditions with the optical density of similar sections taken from livers, the glycogen content of which has been determined by chemical methods. Our results suggest that this method may be relied on to give results accurate to the nearest 0.5% glycogen.

Because of the irregular distribution of glycogen in the livers of animals, the results obtained by this technic can be considered only as indications of general trends, rather than as accurare measurements of the average liver glycogen content. The validity of the observations, however, is indicated by the consistency with which subjects reacted to similar influences. Furthermore, the magnitude of the changes observed was greater than the maximum range of spontaneous variation of distribution.

Normal subjects and patients with various types of disease have been studied. In cases of well controlled diabetes mellitus the liver glycogen was found to be within normal limits. At the onset of diabetic acidosis the liver glycogen was somewhat depressed. In severe acidosis the glycogen levels were reduced to less than 0.5%, but when treatment had controlled the ketosis, the glycogen levels were normal.

Overnight fasts in normal subjects produced only slight and unsignificant depression of liver glycogen levels. In patients starved for periods up to 36 hours a progressive decline of liver glycogen was found. A secondary glycogen rise like that seen in experimental animals has not been observed thus far. The effect of the intravenous administration of small amounts of epinephrine has also been studied. Serial biopsies done before and after breakfast showed an appreciable increase in liver glycogen 30 minutes after eating.

Observations are also being made of the glycogen content of livers with cirrhosis and hepatitis.

Localization of certain spasmodic respirator

responses in the medulla oblongata of the cat. HERBERT L. BORISON (by invitation), GEORGE CLARK and S. C. WANG. *Department of Physiology, College of Physicians and Surgeons, Columbia University.* Systematic exploration of the medulla oblongata of the cat, by means of electrical stimulation, has shown that in addition to gross inspiratory and expiratory responses there may also be elicited a characteristic heightened spasmodic respiratory act which may be likened to such phenomena as coughing, sneezing, gasping or retching. Experiments were carried out on cats under light nembutal anesthesia or on decerebrate preparations. With the aid of the Horsley-Clarke stereotaxic instrument discrete regions of the lower brain stem were stimulated with a bipolar enameled wire electrode connected to a thyratron stimulator (Rev. Scient. Inst. **18**: 669, 1947). Recordings of the respiration were made with simultaneous thoracic and abdominal pneumographs or a spirometer or both. The spasmodic respiratory response could be obtained only on stimulation of the dorsolateral portion of the medulla at the level of calamus scriptorius, which corresponds to the fasciculus solitarius and its nucleus, and also to the general region of the entering glosso-pharyngeal and vagal rootlets. No other region of the brain stem was found to yield similar responses. Attempts will be made to determine the exact conditions which are optimal for specific expression of the spasmodic response in the various respiratory phenomena mentioned.

The effects of anoxia upon myoglobin concentration. WILLIAM J. BOWEN and WILLIAM E. POEL (introduced by HEINZ SPECHT). *Laboratory of Physical Biology, National Institute of Health, Bethesda, Md.* Hurtado, et al (Am. J. Med. Sci. **194**: 708, 1937) reported an increase in myoglobin content of dogs born and reared at high altitudes. Their method did not differentiate between hemoglobin and myoglobin and depended upon perfusion for removal of the hemoglobin. We adopted a method based upon quantitative spectrophotometry of the carbonyl derivatives for the analysis of mixtures of myoglobin and hemoglobin which eliminates the necessity of perfusion. The muscles are cleaned, frozen with dry ice, pulverized and then simultaneously homogenized and extracted with water. The extract is cleared of interfering proteins, buffered and treated with carbon monoxide. Analysis is done at 558 and 538 mμ at which wave-lengths extinction coefficients allow calculation of the concentration of myoglobin and hemoglobin. Using this method, rats, exposed 4 hours daily to 282 mm. Hg (25,000 ft.) are being studied. Body weights, hematocrits and blood hemoglobins are followed as criteria of adaptation. At various stages of adaptation the rats are sacrificed and the gastrocnemius and soleus muscles analyzed. To date, nine rats, exposed for 2 to 52 weeks, have been analyzed. In these the hematocrit and hemoglobin values had increased 25 to 66 per cent over the averages of the controls. The myoglobin, however, consistently shows no significant change from that of the controls. Also, the results show that 2 to 8 times more hemoglobin is retained by the muscles of the exposed animals than by the controls and that the amount retained varies directly with the hematocrit value.

Alkali decomposition of myoglobin. WILLIAM J. BOWEN (introduced by HEINZ SPECHT). *Laboratory of Physical Biology, National Institute of Health, Bethesda, Maryland.* Among the differences reported to exist between hemoglobin and myoglobin is greater resistance of myoglobin to alkali decomposition. Haurowitz (Z. Physiol. Chem. **232**: 125, 1935) gives data showing that 14 hours is required for 86% decomposition of human myoglobin by .05N NaOH and that 38 hours is required for complete decomposition. The same author (Z. Physiol. Chem. **183**: 78, 1929) also shows that the rate of decomposition of hemoglobin by alkali varies according to species. The reactions of metmyoglobin and oxyhemoglobin of the dog and horse with N/10 NaOH have been followed with the aid of a spectrophotometer at 24 ± 1°C. The extinction coefficients of 100% alkaline metmyoglobin and of oxyhemoglobin and of the respective alkali hematins at 540 mμ were used. Four experiments, using each globin in each experiment, were done. The average fractions of myoglobin (Mb) and of hemoglobin (Hb) decomposed at various intervals are given in the table. The results show that myoglobin of both the horse and dog is more resistant to alkali

Time	Per cent decomposed			
	Horse		Dog	
	Mb	Hb	Mb	Hb
15 sec.	18.8	17.6	35.6	93.1
1 min.	23.9	25.0	43.3	92.6
5 min.	28.2	56.1	53.0	88.3
1 hour	36.8	94.9	60.8	87.5
24 hours	69.7	95.8	96.5	93.2

decomposition than their respective hemoglobins. They also show that the rate of decompositon of myoglobin by alkali differs in the two species.

Enzyme inhibitors on conduction and respiration of frog nerve. L. L. BOYARSKI, S. POSTEL, and A. ROSENBLATT (introduced by R. W. GERARD). *Dept. of Physiology, The University of Chicago.* Sodium fluoracetate, 0.001M to 0.1M (over a pH range of 5 to 8) does not affect conduction or resting respiration of frog sciatic nerve (summer frogs). Methylfluoracetate, 0.005M, which should penetrate more readily, irreversibly abolished the action potential in about 3.5 hours. Extinction is faster at higher

concentrations, about 2 hours at 0.05 M. At about two hours, the action potential has been reduced 50%, but resting respiration is already inhibited by 80%. This latter differs from muscle, where only activity metabolism is affected (McKeen Cattell; Cori et al, Report OSRD 4984, June 2, 1945). Fumarate, 0.01M, applied simultaneously with, or 30 mins. after, the poison, completely protects the action potential while permitting a fall of 50% in resting respiration. Acetate, alcohol, pyruvate, or succinate, at 0.01M, do not prevent the fall in action potential, though 0.05M succinate protects. Other substrates are being tested at higher concentrations, and the study is being extended to respiration of stimulated nerve. With the Gerard-Tobias micro-respirometer, using 3 mg. samples, the Q_{O_2} for left and right nerves agree within 10%. The coefficient of variation among all frogs (of species Rana pipiens) was 20% (in 32 experiments). At 30°C., Q_{O_2} during the first half-hour is 140 ± 28.

The myenteric reflex. EMIL BOZLER. *Dept. of Physiology, Ohio State University, Columbus.* In the dog's small intestine, stroking the mucosa increases the strength of the rhythmic contractions on the oral side. In the rabbit's small intestine, stroking is ineffective but longitudinal stretching causes strong contractions of the circular muscles, also exclusively on the oral side. The contractions arise above the stimulated region but may be conducted in either direction. Local application of nicotine sulfate in concentrations as low as 1:20,000 blocks the spread of the response orally. It is concluded that an enteric synaptic mechanism conducting impulses chiefly or exclusively orally is responsible for the polar responses of the intestine. In the dog, the introduction of a bolus induces, on the oral side, strong rhythmic activity of the same frequency as that of the empty organ. Action potentials are similar to those associated with the activity of the empty intestine. The evidence of this and previous work indicates that the individual contractions are myogenic but that the strength of the contractions can be modified by an enteric reflex mechanism.

The effect of morphine on urinary volume in women. J. T. BRADBURY, O. F. KRAUSHAAR (by invitation), W. E. BROWN (by invitation). *Dept. of Obstetrics and Gynecology, State University of Iowa, Iowa City, Iowa.* This is a clinical report of the effect of morphine on the urinary output in ten (10) women. The patients studied represented various adult ages; and multiple observations were obtained on each patient. Each patient had nothing by mouth from midnight until 3 p.m. on experimental days. A retention catheter was inserted at 7 a.m. and the intravenous administration of fluids begun. In each case 2,400 cc of fluid was given intravenously, and the rate was controlled so that the total volume was introduced into the vein in approximately 5 hours. The experiments were done on days when the patient was afebrile and clinically seemed to be in water balance. Hourly urines were collected for 8 hours and the volume and specific gravity determined. The urine for the next 16 hours was collected as a single specimen. On the test days, Morphine Sulfate gr. ¼ I.M. was given to each patient at the time the intravenous fluids were begun, regardless of age or weight.

The hourly urine volumes were markedly diminished after morphine was given so that the average 8-hour excretion volume was one-half that on control days; 890 cc as compared to 1770 cc. The urine flow increased 8 or 9 hours after the morphine was given so that the total excretion in 24 hours was similar on control and morphine days.

The secretory pressure of the liver of the chicken. N. R. BREWER (introduced by A. B. LUCKHARDT) and R. L. GUNNING. *Dept. of Physiology, The University of Chicago.* This report covers sixteen recordings of the bile pressure from the hepatic duct. The maximum pressures obtained were: 253, 152, 165, 140, 125, 190, 119, 175, 147, 186, 160, 172, 180, 190, 173 and 167 mm of water. This gives an average maximum pressure of 168 mm of water and a range of 119 to 253 mm of water. Rhythmic fluctuations of pressure were noted in all cases. These fluctuations were from 10 to 30 mm of water and occurred everyone to five minutes. Upon observation of the hepatic duct it was found that this pressure variation was due to peristaltic action along the duct. After the bile pressure reached the above peaks, a gradual decline in pressure was observed. The first nine experiments were made on chickens anesthetized with sodium barbital. The last seven chickens were anesthetized by positive pressure ether insufflation. In the first nine experiments the hepatic duct was cannulated about a centimeter from its entrance to the intestine. In the last seven experiments, besides cannulating the hepatic duct as in the first experiments, the small connecting duct between the liver and the gall bladder was clamped off with a hemostat close to the liver.

Thermodynamic analysis of the relative effectiveness of narcotics. F. BRINK and J. M. POSTERNAK (by invitation). *Johnson Foundation, University of Pennsylvania.* In a multiphase system in equilibrium with respect to a particular constituent its thermodynamic activity in experimentally inaccessible phases equals that measured in another phase. This principle is useful in analysis of chemical effects on living cells where only concentrations in extracellular phases are known. Since narcosis is a process apparently reaching equilibrium this method is applicable to systematic study of the relative effectiveness of narcotics. Ferguson (Proc. Roy. Soc. *B*, **127**: 387

1939) reported that in contrast to greater than thousandfold variation in aqueous concentrations of equally effective doses of many substances the corresponding thermodynamic activities lie in a relatively narrow range of values. The present calculations show that for many narcotics of different chemical structure, acting on several cell types, equal degrees of effect are produced at approximately equal thermodynamic activities. This conclusion contrasts sharply with findings of Ferguson and of Badger (Nature, **158:** 585, 1946) both of whom emphasize the progressive increase, in an homologous series of compounds, of the thermodynamic activity at which narcosis occurs. It seems that both of these systematic relations among the activities of narcotic substances can occur. (In these calculations the pure narcotic at the same temperature is chosen as the standard state.)

New experimental evidence to be presented confirms the existence, for some cell types, of the law that equal degrees of narcosis are produced at equal thermodynamic activities.

Circulatory reserves shown by animals under acceleratory exposure. S. W. Britton, and V. A. Pertzoff (by invitation), *University of Virginia Physiological Laboratory.* Reduction in carotid arterial pressure observed during exposure to acceleratory forces may be considered as a blood pressure deficiency area in time. Such a deficiency area is directly proportional to the product of the intensity and duration of centrifugation. Used in correlation with post-acceleratory blood pressure changes, a significant evaluation of an animal's circulatory reserve condition may be obtained.

Monkeys and dogs were tested at $\frac{1}{4}$ g. for 5–30 sec. Of the two factors involved, animals appeared to be more sensitive to intensity than to time of exposure to g forces. Comparison of the effects of fasting, exercise, cold, anesthetics, hemorrhage, operative and other conditions was made with the responses of the normal animal. Marked deficiencies in recoverability were observed by the circulatory index method, even after mild acceleratory exposures only were employed.

Food intake as a mechanism of temperature regulation in rats. John R. Brobeck. *Laby. of Physiology, Yale Univ. School of Medicine, New Haven, Conn.* That food intake varies with environmental temperature is a widely accepted principle; and it is generally assumed that this relationship is derived from the fact that total energy expenditure varies with temperature. Thus, food intake is thought to be determined by total energy expenditure. The following experiments suggest, however, another interpretation. Adult male rats were acclimatized to 84°F., and then exposed for 18 hours to any one of several temperatures ranging from 65° to 97°. Food intake, water intake, weight change and body temperature change were measured. At the lower temperatures (65°–72°F.), food intake increased, and body temperature rose by about 1.0°F. (presumably because of the animals' greater alertness and activity at the end of the exposure period). A similar rise in body temperature was observed at the other end of the temperature scale, i.e., at 94°F., but here the food intake was extremely low, and the animals lost weight because their expenditure was so much greater than their energy intake.

At these two levels (70° and 94°F) body temperature change was the same and total energy expenditures were comparable; yet food intake was entirely different. These data suggest that under the conditions of these experiments food intake served as a mechanism of temperature regulation.

The production of waves of inhibition in the esophageal-fundic region. Daniel A. Brody (by invitation), J. D. Lawson (by invitation) and J. P. Quigley. *Department of Physiology, University of Tennessee, Memphis, Tennessee.* In Bayliss and Starling's original demonstration of the Law of the Intestine the presence of a relatively large foreign body in the intestine was essential to the production of the wave of inhibition preceding the wave of contraction. Similarly, we have observed that deglution regularly produced a transient fall of intralumenal pressure within both the fundus and lower esophagus in 8 out of 10 human subjects (74 of 78 trials) when the pressure recording device was in these two regions. However, when recording tips were only in the stomach, deglution produced no significant pressure change in the body and fundus. In the former situation, the esophageal pressure (measured by the open tube-optical manometer technic of Brody and Quigley, Gastroenterology, In Press) began to fall 0–3 seconds after the onset of swallowing, fell 5–15 cm. of water within 4 seconds and rose to normal in approximately 10 seconds. This negative pressure wave was immediately followed by an elevation of pressure in the esophagus to 10–35 cm. of water above the basal level and then returned to normal in 6–9 seconds. The negative pressure wave 22–45 mm. distally in the fundus began 0–2 seconds after the onset of the esophageal negative wave, fell 3–12 cm. of water, persisted 10–14 seconds, then returneb to normal. Succeeding positive waves did not exceed 5 cm. of water in amplitude. Perhaps the wave of inhibition preceding the wave of contraction is a reserve mechanism elicited when the contraction wave alone is inadequate to propel a bolus.

Measurement of gastric tonus in the normal human. Daniel A. Brody (by invitation) and J. P. Quigley. *Department of Physiology, University of Tennessee, Memphis, Tennessee.* Gastric tonus (the tension exerted by a given number of muscle fibers, Brody and Quigley, Bull. Math.

Biophysics, In Press) was determined in five normal human subjects. The method consisted of (a) recording the intraluminal pressure (Brody and Quigley, Gastroenterology, In Press) of the 6 hour fasting stomach distended to a standard volume of 600 cc., (b) measuring the pressure decrease following the administration of three drops of nitroglycerine sublingually or amyl nitrite by inhalation until severe head throbbing developed, (c) solving the equation T = Pr/2, where T = wall tension/cm. (tone), P = fall in pressure and r = 5.24 cm, the radius of a 600 cc. sphere. The distended stomach is not entirely spherical and its wall is not uniform, thus the calculated values of tone are approximate. However, this method appears to supply the most accurate measure of tone under physiological conditions so far developed. The fall in basal intragastric pressure occasioned by nitrites was 3.0–7.3 cm. of water; corresponding to a reduction of tone of 7.3–19.1 gm. Assuming the complete abolition of tone by nitrites, then the original gastric tone was 7.3–19.1 gm. In general, the magnitude and duration of tone reduction was inversely proportional to the degree of gastric phasic activity which preceded the nitrite administration. In one completely vagotomized subject, exposed to 18 hours constant Wangensteen drainage, nitrites lowered gastric tone 14.8 gm.

Insulin inactivation by tissue extracts. R. H. Broh-Kahn (by invitation) and I. Arthur Mirsky. *May Institute for Medical Research of the Jewish Hospital, Cincinnati, Ohio.* A system capable of inactivating insulin during *in vitro* incubation has been demonstrated in rat, rabbit, beef and human livers. In the rat, this system has also been found in smaller amounts in the kidney and in muscle; plasma appears to contain none. The muscle of the rabbit is a poorer source than rat muscle. The insulin inactivator is extracted after homogenization of the freshly excised tissues in 3 volumes of ice water and is found in the supernatant fluid from the centrifuged homogenate. The active fraction can be precipitated by adjustment to pH 5.0 and partially recovered by redissolving at pH 7.5. The inactivator from rat liver is believed to have the properties of an enzyme. It is destroyed by heating to 70° C. for 10 minutes. Its maximum activity during incubation is demonstrated at pH 6 to pH 8. The effect of the temperature during incubation has not been thoroughly investigated although activity is observed at 37° C. and not during incubation at 5° C. At 37° C. and pH 7.5 the destruction of insulin proceeds at a fairly constant rate for the first part of the incubation and then decreases progressively. Dialysis of rat liver extracts results in a loss of activity. The activity of such a preparation can be recovered by the addition of magnesium or manganese salts. The activity of an undialyzed extract can be almost completely inhibited by low concentrations of cupric and zinc salts but is not affected by salts of magnesium, manganese, calcium or sodium. 10^{-3} M iodoacetate and iodosobenzoate also inhibit its activity. This inactivator is not believed to have the properties of known proteolytic enzymes since the extracts do not produce appreciable splitting of a hemoglobin substrate at pH 7.5 nor is their activity inhibited by the relatively non-specific protease inhibitors of the soybean.

The sequence of functional changes in a neurone during narcosis and anoxia. D. W. Bronk, F. Brink and M. G. Larrabee. *Johnson Foundation, University of Pennsylvania.* It is the purpose of this communication to consider how a number of related characteristics of nerve are modified by narcosis, anoxia and certain ions.

Thus, if chloretone be applied to frog axons, a series of changes ensue with respect to time and concentration. Under low concentrations there is first the well known rise in threshold. Under higher concentrations a still greater stability of the nerve is revealed by its inability to initiate rhythmically recurring impulses under the stimulus of a chemical agent such as sodium citrate. This region which has lost the power to initiate impulses can however conduct impulses which originate in another portion of the nerve. A still higher degree of stability, induced by higher concentrations of chloretone, are necessary to abolish conduction. Analogous, perhaps, is the observation that synaptic excitation in a ganglion is blocked under lower concentrations than is axonal conduction.

Further evidence of these progressive alterations in the properties of nerve are found in the changes in oxygen utilization: in the time course of the extra oxygen consumption of activity and in the resting oxygen consumption.

The inter-relation of these several functional characteristics of nerve which are thus progressively modified will be considered in terms of the relative actions of anoxia and various ions as well as of narcotics.

Antidromic potential recordings from the medullary pyramid of the cat. John M. Brookhart and Russell E. Morris (by invitation). *Institute of Neurology, Northwestern University Medical School, Chicago, Ill.* As a foundation for projected studies of the pyramidal system, recordings have been derived from the medullary pyramids of cats anesthetized with Dial or anemically decerebrated. Antidromic activity has been initiated in these fibers by stimulating electrodes placed in the region of the lateral cortico-spinal tracts at various cord levels. Sharply localized pickup from the medullary pyramids is possible using a monopolar needle electrode oriented in a Horsley-Clarke instrument.

Due largely to the impossibility of exciting all

cortico-spinal fibers simultaneously, the resultant recordings are not strictly uniform since the active fibers vary in distance from the recording electrode. Features common to all recordings include: (1) a prominent deflection initiated at an indicated conduction velocity of approximately 35 m.p.s., reaching a peak at an indicated velocity of 14–20 m.p.s.; (2) one or more subsequent elevations reaching their peaks at indicated velocities between 10 and 5 m.p.s.; (3) a total duration of discharge varying between 10 and 20 ms. Sporadically, small elevations are seen having indicated velocities of 85–100 m.p.s. and 40–50 m.p.s. Indications of the slowest fiber responses are obtained only with conduction distances of less than 50 mm. It has thus far been impossible to activate the pyramid antidromically with cord stimuli applied caudal to the upper thoracic levels. Fiber diameter measurements indicate that the great mass of fibers lies in the 1.0–4.0μ range with only scattered fibers exceeding 5μ, the largest measured being 11μ in diameter.

Responses of inhibited motoneurones. CHANDLER MCC. BROOKS, J. C. ECCLES (by invitation) and J. L. MALCOLM (by invitation). *From the Departments of Physiology, The University of Otago, Dunedin, N. Z., and the Johns Hopkins University, Baltimore, Md.* Inhibition of reflex discharge of cat's gastrocnemius or quadriceps motoneurones has been effected either directly by a centripetal volley in the large proprioceptor fibres of the antagonistic muscles or more indirectly by a volley in other afferent nerves of that limb (proprioceptive or cutaneous). The testing responses of such motoneurones have been evoked either by monosynaptic excitation (the myotatic reflex) or by volleys fired antidromically along their motor axons.

(i) Inhibition has no appreciable action on the initial rate of rise of the pure synaptic potential (recorded either from ventral root or by focal electrode), but it lowers the summit and expedites the decay. (ii) Furthermore inhibition increases the critical voltage at which a synaptic potential generates a reflex discharge. (iii) Motoneurones may be deeply inhibited and yet show no depression of the propagation of antidromic impulses into their somas, i.e. of the soma spike potential (confirming Renshaw). (iv) Nevertheless inhibition depresses the facilitation of the soma spike potential that occurs during a synaptic potential (Brooks and Eccles, J. Neurophysiol., 1947, **10:** 251), this depression paralleling effect (i) above.

These observations accord well with the hypothesis that inhibition is produced by focal areas of anelectrotonus on the motoneurone's soma (Brooks and Eccles, Nature 1947, **159:** 760). Such areas would limit the spread and fusion of the local responses set up under the excitatory synaptic knobs, and hence produce effects (i) and (ii). On the contrary, antidromic propagation into the soma will be mediated by currents flowing through widely spreading circuits and so should be little if at all depressed (effect iii) by the postulated anelectrotonic foci, particularly when the complementary dispersed catelectrotonic areas are taken into account. Effect (iv) derives from (i).

Enzyme inhibitors on electric activity of frog brain. V. B. BROOKS, R. E. RANSMEIER, (introduced by R. W. GERARD). *Dept. of Physiology, The University of Chicago.* The electrical activity of the isolated frog brain, both the spontaneous normal rhythm and convulsive-type caffeine spikes (Libet, B., and Gerard, R. W., J. Neurophysiol., 1941, **4:** 438), was utilized to study some relationships between metabolism and brain activity.

The anti - cholinesterase, tetraethylpyrophosphate (TEP) 10^{-7} or 10^{-3} M) markedly increases the amplitude and regularity of the normal 4–6 a sec. rhythm, and maintains this for a long period. (Compare ACH-eserine, Gerard, R. W., Ohio J. Sci. **41:** 160, 1941) Di-isopropylfluorphosphate (DFP) (10^{-2}M), but not TEP, may slow the waves to two a sec. and render them more spike-like. DFP (10^{-2}M), but not TEP (10^{-3}M), completely suppresses spikes otherwise obtained with caffeine alone. The metabolic inhibitor, methylfluoracetate (0.05M), depresses the amplitude of the spontaneous rhythm, but leaving the frequency unchanged, and reduces the period of surviving activity of the excised brain to about one-tenth of normal. Methylfluoracetate, like DFP, fully suppresses caffeine spikes. Sodium fluoracetate (0.05M) has no significant effect on the spontaneous or caffeine-induced activity; presumably because of poor penetration (see abstract Boyarsky, et al.). Experiments with respiratory intermediary compounds will be discussed.

The pulmonary arterial pressure of the chicken F. BROWN (by invitation) and S. RODBARD. *From the Cardiovascular Department, Research Institute, Michael Reese Hospital, Chicago, Illinois.* We have recently investigated the nature of the body temperature-systemic blood pressure relationship which is seen in frogs, turtles, mammals and birds. In these animals a lowering of the body temperature results in a fall in arterial pressure; increasing the temperature results in a return of the pressure to normal.

The pulmonary arterial pressure of the turtle, being essentially the same as the systemic arterial pressure, also varies with the changes in body temperature. It was therefore decided to determine if similar changes occurred in the pulmonary arterial pressure of a warm blooded animal as body temperature was varied. The normal pulmonary arterial pressure of the chicken was found to average

20/8 mm. Hg. This is approximately the pulmonary pressure reported for mammals. The warming or cooling of the chicken had no effect on the pulmonary pressure although the systemic arterial pressure did change significantly. Neither acetylcholine nor epinephrine had any significant effect on pulmonary arterial pressure, as in the mammals. It would thus appear that the temperature-pressure relationship does not apply to the pulmonary arterial circulation in the bird. Furthermore, these results show that the pulmonary arterial pressure remains at about the same level in warm blooded as in cold blooded vertebrates while the systemic arterial pressure rises considerably above that in cold blooded vertebrates.

Effect of tetra-ethyl-ammonium chloride on intra-arterial blood pressure in patients with coarctation of the aorta. GEORGE E. BROWN, JR. (by invitation) and EARL H. WOOD, *Section on Physiology of the Mayo Clinic and Foundation, Rochester, Minnesota.* Intraradial and intrafemoral arterial pressures of twelve patients with coarctation of the aorta were recorded continuously by hypodermic strain gage manometers. Simultaneous records of ear pulse, blood content of the ear, heart rate, respiration, and saphenous venous pressure as well as electrocardiograms, were obtained, in most instances. Continuous records were obtained with the persons in the supine position and during tilting to the 65 degree upright position for periods of fifteen to one hundred twenty seconds. Tetra-ethyl-ammonium chloride was injected intravenously in doses of 4.4 to 7.7 mg. per kilogram of body weight. Continuous records were obtained with the patients supine and during intermittent tilting for periods up to sixty minutes after injection of the drug. After injection of the drug with the patients supine, maximal decreases of intraradial pressure below average values obtained in the control period averaged 36 (12—69) mm. of mercury, systolic, and 8 (−1–19) mm. of mercury, diastolic. Intrafemoral pressure decreased an average maximum of 22 (6 – 38) mm. of mercury, systolic, and 6 (−3 – 20) mm. of mercury, diastolic. Postural hypotension was observed in these patients after administration of the drug. In seven normal persons studied under similar circumstances, the average maximal decrease in radial systolic pressure produced by the drug was 24 (14 – 36) mm. of mercury. The average decrease in diastolic pressure was 1 (−12 – 14) mm. of mercury. Figures in parentheses are extreme values.

A method of assaying the efficacy of resuscitating procedures. MARSHALL BRUCER (by invitation) and H. G. SWANN. *Dept. of Physiology, University of Texas Medical School, Galveston.* Any method of assay of a resuscitating procedure demands an accurate prediction of the proximity of death. Among all methods studied—blood gas analyses and respiratory and cardiac behavior—only a characteristic collapse of systolic pressure presaged imminent death with accuracy. Employing optical recording of arterial blood pressure which could be continuously inspected, various resuscitating procedures were applied at different points during the collapse of systolic pressure.

In anoxia from breathing 1% CO in air, rhythmic positive pressure insufflation of the lungs with O_2 was superior to both air insufflation and manual artificial respiration. In anoxia from breathing N_2, insufflation with 5% CO_2 in O_2, or the Emerson "Resuscitator" using O_2, were superior to manual artificial respiration. In anoxia from breathing 2.43% O_2 in N_2, insufflation with O_2 or with 5% CO_2 in O_2, or with the Emerson "Resuscitator" were all superior to manual artificial respiration.

In each of the three types of anoxia studied, resuscitation was successful if begun early but failed if begun late in the period of systolic collapse. There was an intermediate zone during which heart and respiration recovered but only with residual dementia in the animal. The time span between early and late collapse is a matter of seconds—so short that unless resuscitation machinery is immediately available, in practical emergencies the manual method is probably as efficacious as any so far studied.

The uropepsin output in cats given histamine-caffeine in beeswax. GLADYS R. BUCHER and ARTHUR ANDERSON. *Department of Biochemistry, Vanderbilt University, Nashville.* The purpose of this investigation was to learn if uropepsin output in cats is related to gastric hypersecretion stimulated by the daily administration of histamine-caffeine in beeswax. Two series of experiments were performed. These differed in diet and histamine dose. In the first, the diet contained a 9% protein, the daily drug dose was 150 mg. caffeine (alkaloid) with 0.5 mg. histamine base (di-phosphate). In the second there was about 19% protein and 150 mg. caffeine with 0.75 mg. histamine base (dihydrochloride). In the first study, no difference in the average 24-hour uropepsin output of 19 control days and 14 test days was found. One cat that died in convulsions had developed a gastric ulcer. Three months later the cats were used in the second study. After 10 days on the low protein diet and 9 days on the high protein diet, the injections were resumed for 22 days. The uropepsin output was significantly increased in all cats following the change to the high protein diet. In the first eight days of the dose period, all cats increased their outputs still further. In two the increase was statistically significant. Only one of the cats maintained the significantly higher output, the other sickened, refused its food, and the uropepsin output fell off. It appears that uropepsin output is related more closely to the protein consumption

of the animal than to the stimulation of gastric secretion by the use of histamine-caffeine mixture in beeswax.

Consciousness and unconsciousness during anesthesia in relation to brain potential. W. E. BURGE. *Department of Physiology, University of Illinois, Urbana.* When one platinum electrode was placed on the forehead and another on the forearm with a galvanometer connected in the circuit the beam of light was deflected on the galvanometer scale in such direction as to indicate that the forehead was positive to the forearm in 1680 subjects, principally University students and faculty members. Similarly, the forehead was found to be positive to the forearm in 174 hospital patients prior to anesthetization and operation. During anesthetization scalp potential was decreased to zero. In deep anesthesia (plane 3) polarity was reversed in most of the patients, that is, the scalp became negative to the forearm. Upon recovery from anesthesia polarity was again reversed and the forehead became positive to the forearm.

It has been shown (Anesthesiology, Vol. 6, No. 1, 1945) in dogs that the positive potential of the scalp fluctuated with the negative potential of the underlying brain cortex so scalp potential may be used as an index to brain potential. Hence, the positive potential of the scalp observed in the conscious human indicated that the underlying brain cortex was negative; the decrease during anesthesia indicated diminution in brain potential; and the reversal in polarity in deep anesthesia indicated that the brain cortex had become positive. These observations suggest that consciousness and unconsciousness may depend on electrical condition of the brain cortex, consciousness being associated with a gain of electrons and negative condition, an unconsciousness with a loss of electrons and a positive, or less negative, brain cortex.

Epithelial movements in woundhealing in frog corneas. WILHELM BUSCHKE. *Ayer Foundation Ophthalmic Research Laboratory, Manhattan Eye, Ear, and Throat Hospital, New York.* Epithelial movements described previously by many authors as a primary mechanism of epithelial woundrepair, have been studied in detail on stained whole thickness flat preparations of the cornea in frogs. Pinprick injuries heal in vivo and in vitro with the typical radial orientation of the marginal epithelia seen previously in rats (Friedenwald and Buschke, Jl. Cell. & Compar. Physiol. 23, 95, 1944; Buschke, Fed. Proc. 6, 85, 1947). The latent period between injury and onset of orientation is inversely related to temperature with a temperature coefficient of $Q_{10} = 3$, i.e. lower than in the rat ($Q_{10} = 5$), but still of a magnitude as to suggest the importance of metabolic processes. The temperature optimum of the process is between 5 and 25°, i.e. about 15° below that in the rat. Two types of movement and shaping of the individual cells of the woundmargin are seen. One type consists in the extension of pseudopods into the woundzone which eventually join those of other epithelia. This type of movement is seen in the basal layers around pinpricks and in later stages of the healing of larger injuries. The second type of movement consists in a flattening of the marginal cells tangentially to the woundmargin leading to a microscopically continuous often arcade-like outline of the epithelial sheet; this type of movement is seen in the upper epithelial layers around pinpricks and in the early stages in larger injuries and scratches. Pseudopodial processes are most marked in cells which for some portion of their surfaces are separated by an optically clear space from neighboring cells. This suggests that some correlation exists between the spatial relations of neighboring cells of the epithelial sheet on the one hand, and the mode of movement and shaping the individual cell follows in the healing process at different stages and with different sizes and shapes of the injury on the other hand.

Comparative lipotropic activity of parenterally administered pancreatic extracts in dietary fatty livers. J. F. CANEPA (by invitation) and A. C. IVY. *Dept. of Clinical Science, Univ. of Illinois, College of Medicine, Chicago.* The lipotropic activity of both Dragstedt's and Chaikoff's pancreatic extracts was tested by the parenteral route on adult white rats kept on a high-fat (40%) low-protein (5%) diet. This diet is known to produce fatty infiltration of the liver in these animals when fed for 20 days. The pancreatic extracts were prepared according to the techniques described by the aforementioned authors. Three groups of 10 rats each (5 males and 5 females) were used. The extracts, dissolved in saline, were injected subcutaneously every day in the following doses: Group I—167 mgm. of Dragstedt lipocaic per rat; Group II—30 mgm. of Chaikoff's fraction C27 per rat; Group III—control. After 20 days the animals were sacrificed and the livers removed and analyzed for fatty acids, phospholipids and cholesterol. The average total fatty acids content of the liver of the three groups of animals was found to be 3.88, 15.4, and 15.8%, respectively. Similar though not so striking differences were also noted in the phospholipids and cholesterol values. These results show that Dragstedt's and Chaikoff's extracts, though both active by oral administration in the depancreatized dog, behave differently when injected subcutaneously in the rat. Therefore, we may conclude that the lipotropic principle present in these pancreatic extracts is not the same.

Effect of diet on the in vitro synthesis of alamine by liver. ATTILIO CANZANELLI, DAVID RAPPORT, and RUTH GUILD (by invitation). *Department of Physiology, Tufts College Medical School.* Surviv-

ing liver slices from control rats maintained on a standard synthetic diet formed 7.11 ± 0.26 μg of NH_3N/mg. dry tissue/hr. when incubated at 37°C. for two hours in a Krebs-Henseleit medium containing 0.1 M pyruvate and 0.04 M ammonium carbonate. Livers from rats initially on the standard diet and then fasted for 12, 24, 48, 72 and 96 hours, gave the following values respectively: 6.43 ± 0.91; 1.17 ± 0.15; 1.10 ± 0.14; 2.59 ± 0.65 and 2.69 ± 1.04. When rats were placed on a diet containing no protein, but high in carbohydrate for 24 hours the amount synthesized was 8.08 ± 1.02, and for 72 hours was 8.48 ± 0.57. On a non protein, high fat diet for 24 hours, the livers gave 6.19 ± 0.35. With a diet containing no fat but high in carbohydrate, values of 8.20 ± 0.33; 7.09 ± 0.87 and 6.79 ± 1.13 were obtained for feeding periods of 24 hours, 48 hours and 72 hours respectively. On a diet without carbohydrate but high in fat the figures were 3.28 ± 0.69 and 2.82 ± 0.44 for 24 and 72 hour periods. A value of 3.77 ± 0.41 was obtained on a 24 hour diet containing no carbohydrate but high in protein. It appears that fasting markedly reduces the capacity of liver slices to synthesize alamine and that this reduction is due to carbohydrate deficiency.

A method for direct measurement of rate of oxygen utilization by nerve. F. D. CARLSON (by invitation), F. BRINK, and D. W. BRONK. *Johnson Foundation, University of Pennsylvania.* For studying the Q_{O_2} of peripheral nerve we have developed a respirometer in which fluid of known oxygen concentration is drawn at a constant rate through a capillary tube containing a length of nerve and past an oxygen electrode. Variations in oxygen consumption of the nerve cause variations in the oxygen remaining in the solution which passes the electrode. The Q_{O_2} of the nerve is given by

$$Q_{O_2} = \frac{(i_o - i_n)F}{k \cdot w} \frac{\text{moles}}{\text{gm.hr.}}$$

where k is: oxygen concentration sensitivity of the electrode; i_o and i_n electrode currents for empty and nerve filled chamber respectively; W: nerve weight and F: flow rate. Using respirometers having k values of 10^3 microamperes per molar solution of O_2 and a flow rate of 2.2×10^{-4} liter/hr. the change in Q_{O_2} (2×10^{-7} moles/gm. hr.) accompanying 10/sec. stimulation of 9 milligrams of frog sciatic nerve is readily detected. By means of a "bucking circuit" the useful Q_{O_2} sensitivity can be increased tenfold, the limit being set by stability of flow rate and electrode sensitivity.

Electrode linearity and sensitivity must be established for each experimental solution. Slow drifts in sensitivity can be minimized by periodically interrupting electrode current.

Advantages of this respirometer are: continuous recording, scale reading proportional to Q_{O_2}, facility of changing solutions, ease of making functional measurements on nerve, maintenance of constant chemical environment.

An apparatus for the measurement of pulmonary function. L. D. CARLSON, A. W. MARTIN, and V. GATTONE (by invitation). *Department of Physiology and Biophysics, School of Medicine, University of Washington, Seattle 5, Washington.* An apparatus has been assembled for determining alterations of normal pulmonary ventilation in terms of rate of flow, volume of flow, pulmonary emptying rate, residual and reserve air, and oxygen consumption. A flow-meter using a 400-mesh screen as an orifice and recording through a Statham strain gage is interposed between the patient and a Sanborn metabolism apparatus. Gas samples for a Lilly nitrogen meter are drawn from the mouthpiece. Records are made on the kymograph drum and a recording oscillograph. A special attachment at the mouthpiece allows the entire metabolism apparatus to be filled with 100% oxygen. By taking simultaneous flow, nitrogen concentration, and metabolism records, the degree of pulmonary mixing can be seen in the first three breaths, and residual plus reserve air calculated after two minutes. Subsequent measurement of reserve air and other lung volume components complete the data for description of a large number of the phenomena involved in respiration. The apparatus may be used in assessing the value of treatment of asthma. The apparatus may be adapted to bronchospirography (Jacobdeus, *et al.*). The degree of correlation with other methods has not been completely worked out, but the accuracy is adequate for general clinical and experimental tests.

A method for studying reflex activity under varying conditions. L. D. CARLSON and A. W. MARTIN (with design in electronics by R. S. Bark). *Department of Physiology and Biophysics, School of Medicine, University of Washington, Seattle 5, Washington.* This report describes: (1) a simple apparatus suitable for classroom use for classroom use for eliciting a stretch reflex, and recording electrically the isometric responses of the muscle, by means of strain gauges, together with the summated action potentials in the muscle or nerve supplying it; (2) preliminary experiments on the reflex response of a soleus muscle of the cat to stretch and ipsilateral peroneal nerve stimulation under varying conditions. A standardized procedure was used to evaluate the effects of a variety of conditions, *e.g.*, drug action, on reflex arcs of increasing complexity. Muscle tension was recorded from the soleus muscle of decerebrate and spinal cats and muscle action potentials were recorded with coaxial or bipolar electrodes. A stretch reflex was elicited in decerebrate and spinal cats

with a 3 mm. stretch during one second. In decerebrate animals, pentothal in remarkably small doses (0.0026 gm./kg.) abolished the reflex. The convulsant barbiturate (0.0004 gm./kg.) first augmented then depressed the reflex. In spinal animals (24 hours transected) the myotatic hyporeflexia made analysis difficult. Again, the convulsant barbiturate first augmented and then depressed the stretch reflex and induced spontaneous rhythmical activity. Stimulation of the peroneal nerve (50/sec.) inhibited the stretch reflex and induced a post-stimulatory rebound in the decerebrate, but not in the spinal animal. The rebound (ipsilateral extension) is augmented by pentothal (0.0013 gm./kg.), and then depressed (0.00625 gm./kg.). The convulsant barbiturate augmented the effect at 0.0004 gm./kg.

Cell fractionation and gonadotrophin assays of anterior pituitary glands. HUBERT R. CATCHPOLE. *Dept. of Pathology, Univ. of Illinois, College of Medicine, Chicago.* Anterior pituitary glands of sheep, collected on ice, were subjected to cell fractionation by various methods. Following grinding in a mortar with 5 volumes of saline at pH 9.0 (Claude), cell debris and nuclei were removed by centrifugation at 1500 g. (repeated 3 times). Large granules were separated from the extract by centrifugation at 18000 g. for 5 minutes (repeated 3 times) and a microsomal fraction obtained by centrifugation of the remaining supernatant fluid at 18000 g. for 90 minutes. All operations were conducted at 0°C. Satisfactory preparations of nuclei were obtained from the original precipitates by washing and sedimentation. All fractions were also obtained by breaking the cells in a Waring Blendor at a slightly acid pH (6.0) as described in the method of Dounce. Gonadotrophin assays were made on fractions, washings and supernates, using immature rats and mice. Little or no activity appeared in the final supernate by the 'alkaline' method, whereas the 'acid' method extracted about ⅓ of the total measured activity. In both preparations, gonadotrophin activity was heavily concentrated in the large granule fractions. Based on a given equivalent of fresh tissue, microsomes contained only a small fraction (<1/10 – <1/50) of the activity of large granules, possibly representing contamination with active particles of an intermediate size. Nuclei showed a low activity, compatible with contamination by adherent large particles.

The distribution of response thresholds in studies of insect chemoreception. L. E. CHADWICK and V. G. DETHIER (by invitation). *Medical Division, Army Chemical Corps, Army Chemical Center, Maryland, and the Johns Hopkins University.* Data are presented to show that the distribution in an insect population of acceptance and rejection thresholds for chemicals is normal with respect to the logarithm of concentration. The significance of this observation is discussed in relation to the planning of experiments and the interpretation of results.

Changes in blood level of citric acid and calcium in nephrectomized dogs. T. S. CHANG (by invitation) and SMITH FREEMAN. *Department of Experimental Medicine, Northwestern University, Chicago, Ill. and Division of Experimental Medicine, Mayo Foundation, Rochester, Minn.* Blood levels of citric acid and calcium were studied before and after bilateral nephrectomy in dogs. Before the operation the average value of citric acid in the plasma was 5.25 mg. per cent while that for serum calcium was 11.5 mg. per cent. Blood was analyzed every 24 hours after the operation. It was found that both constituents showed an increase; an average of the 24-hour values for citric acid and calcium were 20.3 mg. per cent and 13.52 mg. per cent, respectively. The average values on 5 dogs 48 hours after nephrectomy were 24.5 mg. per cent for citric acid and 12.76 mg. per cent for calcium. Forty-eight hours after nephrectomy, 40 mg. of citric acid per kilo body weight was injected intravenously drop-wise in one hour. Blood samples were taken immediately after injection as well as one, three, and twenty-four hours later. The average values for citric acid and calcium are shown in the following table:

	End of injection, mg. %	1 hour after, mg. %	3 hours after, mg. %	24 hours after mg. %
Citric acid	45.4	42.8	50.1	39.9
Calcium	14.10	14.0	15.0	12.7

Citric acid remained high but calcium decreased to 10.45 mg. per cent before the death of the animals. The dogs survived from 70 to 165 hours after nephrectomy; the average survival period was 114 hours.

The effect of CO_2 upon the ventilation response of the beaver. JOHN L. CHAPIN (by invitation) and HERMANN RAHN. *Department of Physiology, University of Rochester School of Medicine and Dentistry, Rochester, New York.* In his review of the physiology of the diving mammals Irving has shown that in general these animals have a very slight ventilatory response to CO_2 compared to non-diving mammals. The inhalation of 10% CO_2 by the anesthetized beaver is reported as having a very slight effect. In order to rule out the possible depressing effects of the anesthesia experiments were conducted on a 12 kg unanesthetized beaver. A special mask was built and the ventilation, expired CO_2 and O_2, and breathing rate continuously recorded. The minute ventilation during the control period was compared with the 6th–10th minute after breathing the various CO_2 mixtures started.

The ventilation increase was as follows: with 3.8% CO_2 in air—27%; with 6.5% CO_2 in air—84%; and with 11.4% CO_2 in air—252%. These values are somewhat lower than those obtained on unanesthetized dogs in this laboratory, and differ considerably from those obtained on man.

Effect of exercise on renal plasma flow. CARLETON B. CHAPMAN (by invitation), AUSTIN HENSCHEL, JOHN MINCKLER and ANCEL KEYS. *Laboratory of Physiological Hygiene, University of Minnesota, Minneapolis.* In order to obtain quantitative information on the effect of work on renal plasma flow in the normal subject we have devised a method which permits measurements to be made before, during, and after sustained exercise. The clearance substance employed is para amino hippurate and the analytic method that of Bratton and Marshall. The exercise consists of 2 consecutive 15-minute periods on the treadmill. Three levels of exercise have been investigated: A. 3 mph on the level, B. 3 mph at 5 per cent grade, and C. 3.5 mph at 10 per cent grade. All 3 levels produce a progressive decline in renal plasma flow, and the extent of the decrease is a function of duration and severity of the exercise (table 1):

Level	% decline, 15 min.	% decline, 30 min.	No. subjects	No. tests
A	7.5	17.0	3	6
B	17.6	26.6	9	14
C	20.1	33.2	2	4

Return of renal plasma flow to basal values after cessation of exercise is much slower than pulse and respiratory rates and blood pressure. After the mildest of 3 levels of exercise, return to basal values was incomplete in most subjects after 40 minutes of rest, although the residual deficit at that time was not great. It was progressively greater after the same interval of rest following the other 2, more severe, exercise loads. With the heaviest work load, recovery was still incomplete one hour after termination of exercise.

The role of the vagi in the crossed phrenic phenomenon. P. O. CHATFIELD and S. MEAD (introduced by E. M. Landis). *Dept. of Physiology, Harvard Medical School, Boston, Mass.* The term "crossed phrenic phenomenon" refers to the recovery of activity in a hemidiaphragm paralyzed by hemisection of the spinal cord above C3, when the contralateral functioning phrenic nerve is cut or blocked in various ways. Previous investigators have shown that the crossing of descending respiratory impulses can occur on section of the vagi alone. This finding was confirmed during the present experiments performed on rabbits. Crossing was produced also by activation of Hering-Breuer afferents by partial occlusion of a tracheal cannula and by high frequency stimulation of the central end of the cut vagus. Low frequency vagal stimulation inhibited crossing. After bilateral vagal section had produced crossing, an increase in crossing still occurred after inactivation of the functioning phrenic. No afferents inhibitory to crossing could be demonstrated in the cut phrenic. It was concluded that in certain instances crossing may be produced by changes in afferent vagal impulses brought about by inactivation of the functioning phrenic.

Derivation of leads I and III in the dog from analysis of unipolar limb leads. H. M. CHERNOFF (by invitation), W. KAUFMAN (by invitation) and L. H. NAHUM. *Laby. of Physiology, Yale Univ. School of Medicine, New Haven, Conn.* It has been shown that in any unipolar limb lead certain recognized ("proximal") regions of the heart when depolarized produce downward deflections in the electrocardiogram, whi[illegible] other ("distal") regions when depolarized produce upward deflections. Lead I is derived by subtracting the potentials of Vr from those of Vl. It follows that activation of regions of the heart resulting in similar and equal deflections in each of these unipolar leads will not be recorded in Lead I. Such regions are the anterior right and posterior left ventricle; a small portion of upper left anterior ventricle near the septum. Conversely, regions whose activation results in opposite deflections in Vr and Vl will be maximally recorded in Lead I. Such regions are the right posterior and greater part of the left anterior ventricles. Lead III is derived by subtracting the potentials of Vl from those of Vf. Activation of regions resulting in similar and equal deflections in each of these unipolar leads will not be recorded in Lead III. Such regions are right posterior and greater part of left anterior ventricles. Activation of regions resulting in opposite deflections in Vl and Vf will record maximally in Lead III. Such regions are the right anterior ventricle, right and left apex, left posterior ventricle, and small portion of upper left anterior ventricle near septum. This study confirms and amplifies the results of previous experiments on the nature of Leads I and III in the dog.

Ephedrine effect on human uterine contractions. GEORGE P. CHILD (by invitation), B. S. HARDMAN (by invitation), R. A. WOODBURY and R. TORPIN (by invitation). *Departments of Pharmacology and Obstetrics and Gynecology, University of Georgia, School of Medicine, Augusta, Georgia.* The effect of ephedrine sulphate on uterine contractions was studied in normal women and in women with dysmenorrhea at various periods during the menstrual cycle. From 25 to 50 mgm. were given i.v. and the contractions recorded by changes in intra-uterine balloon pressures. There were

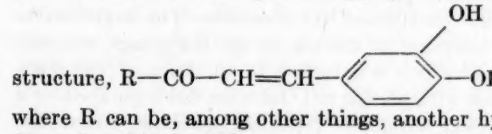
three balloons in the uterus and one in the cervix, each containing one cc. of water. Ephedrine either reduced uterine activity or produced a change in the rhythm of contractions converting disorganized into organized contractions. In the latter case the initial response was the production of one to five slow contractions in the upper segment of the uterus followed by a lower segment contraction wave. The uniform pattern then ensued and was maintained for some time. Many of the patients who were in pain prior to the ephedrine were relieved. The relief of the pain may possibly be attributed to the effect of ephedrine on the central nervous system; yet the uterine effect of the drug must play an important role since pitocin, which has no known C.N.S. effects, influenced the uterus in a similar respect to that of ephedrine and also relieved the pain of dysmenorrhea, see Woodbury, Child, and Torpin these abstracts.

These results, therefore, suggest that the pain of dysmenorrhea may be relieved by drugs which cause or permit the uterus to contract with a uniform rhythmic pattern.

Studies of the mechanism of increased chloride excretion during osmotic diuresis. L. J. CIZEK (by invitation) and J. H. HOLMES. *Department of Physiology, College of Physicians and Surgeons, Columbia University.* The secretion of chloride in dogs was found to be increased during the diuresis produced by the continuous intravenous injection of either 50 per cent sucrose, 50 per cent glucose, 50 per cent sorbital, or 10 per cent urea (12.5 cc. per kilo at 2 cc. per minute). The total amount of chloride excreted varied as the urine volume and bore no relation to the type of diuretic used. Creatinine clearances revealed that as the urine volume increased there was a decreasing reabsorption of chloride from the glomerular filtrate, reabsorption reaching its lowest point at the height of diuresis, returning toward normal as the urine volume decreased. The effect was unrelated to changes in filtration rate and was not modified by the administration of either pituitrin or "doca" (desoxycorticosterone). The same reduction in chloride reabsorption at the height of diuresis was obtained in dogs in which plasma chloride had previously been lowered by osmotic diuretics plus a salt-free diet, and in dogs in which the plasma chloride level was raised during the experiments by infusing a solution containing 50% sucrose and 10% NaCl. It would appear that neither the type of diuretic, the serum chloride concentration, "doca" nor pituitrin modified the tubular activity.

The lack of effect of estrogen on the sex skin of the infant male chimpanzee. GEORGE CLARK. *Dept. of Anatomy, Chicago Medical School, Chicago, Illinois and Yerkes Laboratories of Primate Biology, Orange Park, Florida.* In an earlier study, it was reported that the effects of estrogenic therapy in the male chimpanzee differed from those reported by Gilbert in the chacma baboon. However, the baboons used were young adolescents, while the chimpanzees were young adults, so it seemed possible to explain this difference on the basis of age alone. Although adolescent chimpanzees were not available, it was possible to use two chimpanzees just over two years in age. One had been castrated at the age of ten weeks, while the other was intact. Both animals were given ethinyl estradiol (4.5 mg. per day by mouth for 10 days). This dosage is 180 times that sufficient to produce full tumesence in the female castrate chimpanzee or if measured on a weight basis 900 times the threshold dose. In contrast to reported results for the male chacma baboon but confirming those for the adult chimpanzee no changes in sex skin were observed. There was no swelling of the perianal region, no edema and no reddening of the penis. It may be concluded that a true species difference exists.

Molecular structure and activity of "vitamin P"-like substances. Inhibition of oxidation of epinephrine. WILLIAM G. CLARK and T. A. GEISSMAN (by invitation). *Scripps Metabolic Clinic, La Jolla, Calif. and Dept. of Chemistry, University of California, Los Angeles.* According to the theory of Lavollay, Parrot, *et al* (Helv. Chim. Acta, **29:** 1283, 1946), "vitamin P" acts *in vivo* to prolong the effects of epinephrine. Methods of assay based on this were developed in order to relate molecular configuration to physiological activity. Many flavanones, flavonols, flavones, chalcones and other miscellaneous orthodihydroxyphenolic compounds and metal-complexing agents and their derivatives were examined. The results indicated that high activity depends on the



where R can be, among other things, another hydroxyphenol if the OH groups are placed properly. The ortho-hydroxy groupings must be free. Many compounds were found with markedly greater activity than rutin and "Citrin" (Szent-Gyoergyi). Methylated derivatives such as hesperidin were nearly devoid of activity. Activity probably is related to the reducing powers of the compounds, quinone formation and metal-complexing or chelating capacity. In the light of these and other results it is doubtful that epinephrine antioxidant properties and capillary fragility ("permeability," "Filtration") are exclusively related so far as "vitamin P" effects are concerned.

Effect of tetraethylammonium on responses of isolated intestine to angiotonin and other substances. DEAN A. COLLINS. *Dept. of Physiology, Temple Univ. School of Medicine, Philadelphia.*

Page and Taylor (Science, **105**: 622, 1947) have reported that the tetraethylammonium (TEA) ion increases responses of blood pressure to angiotonin, adrenalin, and usually to histamine, mecholyl, and barium chloride. In the present study the action of TEA on isolated guinea-pig ileum, suspended in Tyrode's solution at 38°C., has been investigated. Angiotonin preparations were made from heparinized dog's plasma incubated with renin and then heated at pH 5. The resulting fluid was treated with tribasic calcium phosphate which adsorbs some impurities. Further purification was effected by adsorption of the angiotonin on charcoal, elution with glacial acetic acid, and precipitation of the angiotonin with ethyl ether. The responses of the ileum to these angiotonin preparations were augmented by the presence of TEA bromide in dilutions from 1 in 13,000 to 1 in 40,000. Responses to histamine were also consistently increased by TEA. Responses to acetylcholine and barium chloride were only occasionally augmented, and were often depressed. That the augmentation of responses to the angiotonin solutions was not due to increased sensitivity to impurities is indicated by the relatively huge doses of control fluids required to elicit responses from the ileum. The control fluids were prepared from plasma by the same procedure used in the preparation of angiotonin except that incubation with renin was omitted.

A method for constant, long-term intravenous infusion of the unanesthetized dog. W. D. COLLINGS, C. J. MARTIN (by invitation) and G. R. WALTERS (by invitation). *Department of Physiology, State University of Iowa, Iowa City.* Apparatus has been designed so that fluids may be infused continuously into a dog's jugular vein for as long as 28 days. The principal parts consist of two brass swivels basically constructed like stopcocks. The larger swivel is mounted in the top of the dog's cage, and the smaller one is attached to the back of the dog's neck with plaster roll. Infusion fluid, pumped by a Brewer automatic pipetting machine, enters the stationary portion of the large swivel. The rotating center portion is connected to the dog by plastic (Tygon) tubing which is attached to the rotating portion of the small swivel. The outlet of the stationary portion of the small swivel leads to 2 mm. (O.D.) plastic (Tygon) tubing which passes subcutaneously to the jugular vein. This tubing is inserted into the lumen of the vein 4 to 8 inches. Thus, with two swivels freely moveable and with a system of heavy rubber bands to keep slackened tubing from wrapping about the dog's neck, there has been little tendency for the animal to become entangled. The equipment can be arranged so that the animal may be removed from the cage for other procedures, such as blood pressure measurement, without any interruption of fluid administration.

Measurements of rapid changes in oxygen consumption by nerve following brief periods of stimulation. C. M. CONNELLY (by invitation) and D. W. BRONK. *Johnson Foundation, University of Pennsylvania.* It has been possible to record an increase in the oxygen consumption of Limulus leg nerves that begins within less than 5 seconds from the start of stimulation. Furthermore, it has been possible to detect an increase in oxygen consumption resulting from the conduction of but a single volley.

The speed and sensitivity necessary for such measurements have been obtained by placing an oxygen electrode (Davies and Brink, R.S.I. **13**, 524, 1942) in direct contact with the nerve. The geometry is such that the oxygen consumption of the resting nerve is balanced by the inward diffusion of oxygen, and the electrode current, which is a measure of the oxygen tension, is steady.

The increased oxygen consumption resulting from the conduction of one or more volleys of impulses causes a decrease in oxygen tension at the electrode. For a brief period of time the change in oxygen tension is a direct measure of the oxygen consumption of the nerve. Thereafter, the oxygen tension is modified by inward diffusion of the gas and the rate of oxygen consumption must be determined by calculation. Accordingly, the method is of especial value for determining the time course of the beginning of the extra oxygen consumption of activity.

Embolization of platelet agglutination thrombi in the hamster's pouch produced by heparin. ALFRED LEWIN COPLEY. *Marine Biological Laby., Woods Hole, Mass. and Laby. of Cellular Physiology, Dept. of Biology, Nen York Univ., New York.* Earlier investigations demonstrated that heparin produces thrombocytopenia in vivo and a decrease of single platelets in vitro. These findings were explained by Copley and Houlihan (Blood, Suppl. 1: 182, 1947) who showed tnat isolated platelets were agglutinated when incubated with heparin plasma. We found that systemic or local heparin injection produced white emboli in the capillary bed of the nictitating membrane of the rabbit. In this investigation we observed the same phenomenon of white thromboembolization in the cheek pouch of the hamster following systemic injections of sodium heparin. This phenomenon was a constant finding in the 32 hamsters studied, and was not observed in the control animals systemically injected with physiologic saline. Immediately following the appearance of these emboli, blood samples were secured directly from the heart, and placed between cover slips. Numerous small white emboli of about leukocyte size were observed in undiluted blood and blood diluted with isotonic sodium citrate solution, 1:10. These emboli which consisted of platelet agglutinates were of the same size

as those in the capillary circulation. Sections of the cheek pouch also revealed, upon Giemsa staining, platelet agglutinates as well as single platelets. Heparin, although highly active as an anticoagulant, does not prevent platelet agglutination, but induces it, which accounts for the thrombocytopenia.

Pilot metabolism and respiratory activity during varied flight tasks. E. L. COREY. *Physiological Laboratory, University of Virginia Medical School.* Determinations of metabolism (103) and respiratory activity (332) were made during varied flight tasks in both a Link trainer and a Piper J-3 airplane. Ten subjects were used, so selected as to represent a variety of flight experience. Caloric output was found to increase with increasing complexity of flight task (straight and level, turns, patterns, rough air) in all subjects, and to be greater in inexperienced as compared with highly experienced pilots. Since these differences were evident even when a specific task demanded no appreciable increase in manipulation of the controls (turns, climbing turns), it was concluded that the raised energy outputs observed represented an increased generalized muscular tension with increased task complexity; further, that this tension was related to pilot experience. The subjects exhibited an increased respiratory rate with an increase in task complexity, although there was considerable individual variation. This respiratory sensitivity to alterations in flight pattern was not, however, invariably correlated with experience. As in the case of caloric output, changes in respiratory rate were not directly correlated with control movements, and the altered respiratory patterns observed were attributed to changing demands in mental activity, resulting in altered, generalized muscular tension.

The results obtained in the J-3 airplane complemented those secured with the Link trainer, and it was concluded that the latter type of study is qualitatively valid for aircraft, with the additional feature of permitting much more rigid control. It was further concluded that determination of pilot caloric output is a valid and convenient indication of the work demand imposed by various flight tasks.

The influence of sodium salts on the extracellular space in experimental hypoproteinemic edema. SAMUEL A. CORSON and ELIZABETH O'LEARY, with the technical assistance of Opal Cain. *Departments of Physiology, School of Medicine, University of Minnesota and Howard University.* Experiments were performed on 3 trained unanesthetized dogs rendered hypoproteinemic by a combination of a low-protein diet and massive plasmaphereses. The plasma proteins fell from an average normal value of 6.6 g. % to an average of 3.4 g. %. The extracellular space was determined by the thiocyanate method (in the postabsorptive state). Repeated administration of NaCl either orally (1 g./kg. body weight) or intravenously (10 ml./kg. of a 1.8, 2.0, or 2.8 M solution delivered by a constant infusion pump at the rate of 2.5 ml./min.) produced no change or led to a decrease in the extracellular space. In only 4 out of more than 20 experiments was there any increase in the extracellular space following NaCl administration. This increase happened to coincide with a significant fall in the hematocrit. Whether there is any causal relationship between the anemia and the change in extracellular space remains to be determined.

Neural control of the renal shunt. J. H. CORT (by invitation) and DONALD H. BARRON. *Laby. of Physiology, Yale Univ. School of Medicine, New Haven, Conn.* Observations on the neural control of the renal shunt indicate that 1) the efferent path is via the splanchnic roots (T10–12); 2) the initial response in rabbits and cats to stimulation of a spinal afferent is unilateral; 3) with continued stimulation the response becomes bilateral in 5–10 minutes in cats, and 3–4.5 hours in rabbits; 4) the crossing occurs in the cord; 5) the anuria which follows an approximation of the crush syndrome in cats can be relieved by novocainization of either splanchnics or the spinal roots T10–12.

Observations on the oxytocic property of human blood. ARTHUR A. COX (introduced by R. R. Overman). *Department of Physiology, University of Tennessee, College of Medicine, Memphis, Tennessee.* It has been reported that blood from patients in labor exerts an oxytocic effect upon the isolated guinea pig uterus, (Compt. Rend. Soc. de Biol. **107**: 88, 1931). We have studied the oxytocic effect of human blood upon the isolated rat uterus suspended in Tyrode's solution in a 37°C. constant temperature bath. Blood from pregnant patients, from males, and from non-pregnant females has been studied. When blood from any of these three groups of patients is added to the uterine bath within five minutes after withdrawal, each cc. of blood is found to exert an oxytocic effect equal to approximately 0.01 units of pitocin. After incubation at 37°C. for one hour, blood from males and non-pregnant females showed no diminution of oxytocic activity, whereas blood from pregnant patients showed a marked reduction of this activity. These results demonstrate the presence of a substance in pregnant blood which is capable of destroying this oxytocic principle. It has been reported previously by others (Am. J. Obst. and Gynec. **52**: 1014, 1946) that pitocin when added to pregnant blood is destroyed by an enzyme, which has been called pitocinase. Blood from males and non-pregnant females does not possess this enzyme. Since pitocinase appears in the blood only during pregnancy and since only in pregnant blood were we able to demonstrate a diminution of oxytocic

activity on incubation, it seems reasonable to conclude that at least a portion of this oxytocic activity is due to the presence of the posterior pituitary hormone.

Analysis of combined effects of exercise and carbon dioxide inhalation on pulmonary ventilation in man. F. N. CRAIG, JANE STUBBS (by invitation) and F. N. MARZULLI (by invitation). *Physiology Section, Medical Division, Army Chemical Center, Maryland.* Normal male subjects inhaled air and mixtures of 2.6 per cent carbon dioxide in air and of 5.0 per cent carbon dioxide in air, from a 600 liter spirometer for periods of three minutes while sitting in a chair, walking at 4.0 miles per hour and running at 5.8 miles per hour on a level treadmill in a room at 20°C. and 40 per cent relative humidity.

The increments of ventilation rate in liters per minute due to either exercise or carbon dioxide inhalation beyond the rate under reference conditions, were augmented by the introduction of the other factor. The ratio of ventilation rate augmented by either factor to the ventilation rate at reference conditions, was diminished by the introduction of the other factor.

The ventilation rate observed when both factors were present together was compared with the rate calculated from the effect of each factor operating alone (1) by adding the increments over the reference condition due to each factor and (2) by multiplying the ratio of exercise augmented rate to reference rate by the ratio of carbon dioxide augmented rate to reference rate:

Calculated rate as per cent of observed ± extreme deviation

Reference Condition	Augmented Condition	Incremental Method	Method of Ratios
sitting, air	running, 5% CO_2	81 ± 5	141 ± 23
walking, 2.6% CO_2	running, 5% CO	92 ± 8	100 ± 11
walking, air	running, 5% CO_2	91 ± 7	105 ± 9
sitting, 2.6% CO_2	running, 5% CO_2	90 ± 4	129 ± 20

The effect of anesthetics on calcium release. MARY CREGAR (introduced by L. V. Heilbrunn). *Zoological Laboratory, University of Pennsylvania.* Experiments were performed in an attempt to investigate the calcium release theory of anesthesia and stimulation. Muscle fibers were removed from frog gastrocnemii and weighed. Half the fibers were immersed for one hour in a solution of 2% ether in frog Ringer and half (controls) were immersed in a frog Ringer solution for the same time. The fibers were then removed and a sample of the immersion fluid was analyzed for calcium. In 9 out of the 10 experiments performed, more calcium was found in the immersion fluid containing ether than in the control fluid. Similar experiments were performed using solutions of 2% cocaine in Ringer and 0.4% chloroform in Ringer. In 7 of the 13 experiments with cocaine, more calcium was found in the cocaine-Ringer solution than in the control, in 4 no difference could be found between the cocaine-Ringer solution and the control, while in 2, more calcium was found in the Ringer solution. Of the 10 experiments performed using chloroform, 7 showed more calcium present in the chloroform-Ringer solution, while in 3 the same amount of calcium was found in both the chloroform and the control solutions. In 6 of the experiments using chloroform the muscle fibers were analyzed for calcium. In 5 of the 6, more calcium was found in the fibers immersed in Ringer than in those immersed in chloroform-Ringer, the sixth showed no difference. In general these results seem to support the calcium release theory.

Errors induced by phosphate in flame photometer analysis of tissue ash for sodium and potassium. J. M. CRISMON. *Department of Physiology, Stanford Univ. School of Med.* In the course of parallel analyses of muscle tissue ash for sodium by the method of Butler and Tuthill (J. Biol. Chem. **61:** 523, 1924) and with the flame photometer, agreement was obtained by the two methods when the dissolved tissue ash was subjected to treatment with CaO for the purpose of phosphate precipitation. Analysis of solutions of tissue ash not subjected to phosphate precipitation yielded values for sodium and potassium appreciably lower than those obtained gravimetrically or with the flame photometer after phosphate precipitation. Additional studies made on muscle ash from rabbits, cats, and rats showed that the presence of phosphate lowered the apparent Na concentration by 24 to 53 per cent and the apparent K concentration by 19 to 29 per cent. Solutions designed to contain 50.0 m. Eq. per liter of Na were made from Reagent Grade or C. P. Samples of NaCl, $NaH_2PO \cdot H_2O$, Na_2HPO_4, $Na_3PO_4 \cdot 12H_2O$, and $Na_4P_2O_7 \cdot 10H_2O$. To each solution was added 10 m. Eq. per liter of K in the form of KCl. Aliquots of each solution were diluted 10 fold, a portion of each was treated with CaO to precipitate phosphates, and the portions, with and without phosphate, were analyzed in the flame photometer for sodium and potassium. For all of the phosphate salts the apparent sodium concentration was approximately 50 per cent of the value measured after the precipitation of phosphate. Potassium concentrations were not altered by the presence of phosphate in the above mixtures. Flame photometer analysis of urine should probably also involve elimination of phosphate.

The effect of inanition on pituitary-adrenal function in the guinea pig. SAVINO A. D'ANGELO, ALBERT S. GORDON, and HARRY A. CHARIPPER. *Department of Biology, Washington Square College of Arts and Sciences, New York University.* A pre-

vious study had indicated that immature guinea pigs fed a normal diet to a body weight of 300–350 grams and then completely starved or chronically underfed display adrenal enlargement characterized by hypertrophy of the inner cortical zones and atrophy of the glomerulosa, thyroid regression, and marked structural changes in the basophiles of the anterior hypophysis (Anat. Rec. **81:** suppl. no. 184, 185, 1941). The present experiments were designed to test, in the starved animal, the generally accepted thesis that cortical hypertrophy is mediated largely through the cortico-trophin mechanism of the anterior pituitary, and that suppression of this mechanism results in cortical atrophy. Female guinea pigs at a body weight of 300–325 grams were placed on a complete starvation regime (water ad lib.) and given daily injections, for 5–6 days, of either 2–5 cc. of a saline cortical extract or 2.5–15.0 milligrams of desoxycorticosterone. It was found that the administration of these cortical substances to the starved guinea pig failed to prevent cortical hypertrophy in animals losing 30–50% of their body weight, and failed to alter appreciably the course of the body weight loss. In contrast, guinea pigs hypophysectomized for 2–9 days with corresponding degrees of weight loss showed cortical atrophy. Further attempts are being made to inhibit the adrenal enlargement of starvation by the administration of much higher doses of potent cortical materials. The significance of the cytological changes in the anterior hypohysis as related to the cortical hypertrophy in starvation will be discussed.

Alterations in the rate of glycolysis in human blood following the addition of salts. T. S. Danowski. *Department of Research Medicine and the Renziehausen Foundation, University of Pittsburgh School of Medicine.* The disappearance of glucose during the incubation of defibrinated human blood at 36 to 37°C. was measured in a series of experiments. The addition of NaCl in amounts sufficient to increase its concentration in blood by 40 meq. per liter had no effect on glycolysis. Additions of $CaCl_2$ which increased the concentration by 40 or by 20 meq. per liter invariably retarded glycolysis, while increments of 2.5 meq. per liter were without effect. Equivalent amounts of KCl or $MgCl_2$ only rarely slowed the disappearance of glucose. Glycolysis was accelerated, on the other hand, by the addition of NH_4Cl producing increments as small as 2.5 meq. per liter. The introduction of HPO_4^-, $SO_4^-\cdot$, and of $HCO_3^-\cdot$ as the sodium salt hastened in each instance the disappearance of glucose. The accelerating effects on glycolysis of the HPO_4^- and the NH_4^+ ions appear to be additive, since glucose disappeared more rapidly from blood which contained added $(NH_4)_2\ HPO_4$ than it did from blood with equivalent amounts of either NH_4Cl or Na_2HPO_4.

The time course of in vivo oxygen consumption of cerebral cortex following electrical stimulation. P. W. Davies (by invitation), R. G. Grenell and D. W. Bronk. *Johnson Foundation, Univ. of Pennsylvania.* The oxygen cathode has been employed to make in vivo measurements of local oxygen consumption at the surface of the cerebral cortex after a period of electrical stimulation. The experiments were performed under moderate Dial anaesthesia on cats with a trephine hole over the supra sylvian gyrus. Oxygen consumptions were taken at intervals by measuring the rate of fall of oxygen tension when the oxygen cathode was suddenly pressed sufficiently hard to occlude the local circulation.

At the end of a strong stimulus (100 shocks/sec., ten seconds duration, 40 v. open circuit at the output of the isolating transformer) the rate of oxygen consumption is usually twice that obtained before stimulation. Within a half minute, however, the rate falls to about two-thirds of the "resting" value. By this time also there is a marked reddening of the venous blood. Afterwards there is a period of recovery during which the oxygen consumption rises gradually to the "resting" rate. This period may be as short as two minutes but is often longer. A much weaker stimulus (4 v.) of the same frequency and duration causes hardly any increase in oxygen consumption, but leads to the same reduction and recovery experienced with the strong stimulation. The relation of these findings to those of Dusser de Barenne and McCulloch on facilitation and extinction, and to those of de Leão on spreading depression, will be considered.

Thermal irritation of alimentary mucosa. Robert E. Davis (by invitation) and A. C. Ivy. *Dept. of Clinical Science, Univ. of Illinois College of Medicine, Chicago.* A subject of particular interest in the genesis of peptic ulcer and carcinoma is the possibility that ingestion of hot substances may irritate the mucosa of the upper alimentary tract. The present study is an attempt to correlate degrees of heat injurious to mucosa with that to which the human subject exposes himself. Temperature tolerance was determined in 120 medical students by measuring the maximum temperature at which they would drink coffee in rapidly successive swallows. The average temperature was 57.8° Centigrade, with a range from 51°C. to 68°C. The standard error for this series was 0.26°C. Subjects with temperature tolerance of 68°C. ingested successivly 50 cc., 100 cc., and 200 cc. of water at that temperature, and intra-gastric temperatures were measured by means of a gastric thermacouple. The maximum temperature, 49.9°C., was reached within ninety seconds, with rapid cooling and return to normal intragastric temperature within 30 minutes. In acute experiments on dogs, gastric, esophageal, and buccal mucosae were bathed with

water at graded temperatures, and biopsies taken. Coagulation and superficial sloughing of gastric mucosa were demonstrated only at 65°C. or above. Similar changes occurred in esophageal mucosa at 60°C. or above. Buccal mucosa blistered and sloughed at the relatively low temperature of 55°C., which is within the range of human temperature tolerance. It remains to be proven whether or not the animal thresholds for thermal injury obtain in the human subject and whether repeated exposure of the stomach to a temperature of 50°C. will cause histological changes.

Influence of intravenous cytochrome c upon visual acuity of the dark adapted human eye. STANLEY K. DAVIS (introduced by J. W. Heim). *Aero Medical Laboratory, Wright Field, Dayton, Ohio.* Mitigation of effects induced by hypoxia, both anemic and anoxic, through parenteral administration of cytochrome c in animals and man has been reported in recent months by Proger and Dekaneas (Science, October 25, 1946, pp. 389–390). No corroborative studies have as yet reached publication, but two failures to duplicate their results in animals (rats) have been reported (Scheinberg and Michel (Science, April 5, pp. 365–366); Stadie and Marsh (J. Clin. Invest. **26**: 899)). No further reports on human use have yet appeared. The present study was designed to determine any effect of intravenously administered cytochrome c (Wyeth) upon the visual acuity of the dark adapted eye under hypoxic conditions. Seven human subjects were subjected in "blacked out" altitude chambers to atmospheric pressures equivalent to 18,000 feet for a total of 35 exposures of one-hour duration each. Each subject was pre-adapted before each exposure until basal visual thresholds were achieved. Thresholds were determined every 10 minutes during exposures with a portable Hecht-Schlar Adaptometer (Cenco) and were recorded as logarithms of the light intensity just sufficient to permit identification of a figure interposed against the light source. Intravenous cytochrome c solutions (50–100 mg.) and colored saline were given alternately prior to exposure of each subject. Subjects were kept ignorant of the content of injections. Oxygen tension in the chamber was checked continuously with a Pauling Oxygen Meter.

With the method described above, it was not possible to demonstrate significant differences in the rate or degree of deterioration in visual acuity in human subjects prepared with and without intravenous cytochrome c (Wyeth).

Adrenaline hyperglycemia in hypophysectomized dogs. R. C. DE BODO, I. H. SLATER (by invitation), H. F. WEISBERG (by invitation), K. F. PRESCOTT (by invitation). *Department of Pharmacology, New York University College of Medicine.* As previously shown, hypophysectomized dogs with adequate amounts of liver glycogen show a smaller hyperglycemic response to intravenous adrenaline than normal dogs. In the present work, the development of this altered response, its relation to insulin hypersensitivity and glucose utilization was studied. The test dose of adrenaline, 0.0035 mg./kg./min. for five minutes intravenously, causes a marked hyperglycemia (maximum rise 43–81, average 58 mg. per cent) within fifteen minutes from the start of the infusion. Four–five days after hypophysectomy the adrenaline hyperglycemia is distinctly decreased and becomes gradually less pronounced as the postoperative time increases. After four weeks the hyperglycemic response is slight. This phenomenon seems to develop parallel with the hypersensitivity to insulin as described by Slater et al in these Proceedings. When the hypophysectomized animal has reached the stage at which the adrenaline hyperglycemia is greatly reduced, the intravenous infusion of glucose produces a marked and prolonged hyperglycemia demonstrating that the abnormality cannot be attributed to increased utilization of the glucose liberated by adrenaline but must rather be related to the deficient mobilization of liver glycogen. Furthermore, when the blood sugar of normal animals is maintained at a low level by large doses of insulin, adrenaline still produces considerable hyperglycemia (maximum rise 31–46 mg. per cent). This is further evidence that increased insulin activity is not the cause of the decreased adrenaline hyperglycemia noted in hypophysectomized animals. The relationship of this resistance to the hyperglycemic action of adrenaline to the various endocrine organs is being examined and will be discussed.

The rod calorimeter. RONALD DEERING (introduced by A. B. HERTZMAN). *Department of Physiology, St. Louis Univ. Medical School.* The rate of transfer of heat from deeper tissues to a limited area (1 cm^2) of skin surface may be estimated by means of a specially designed rod calorimeter. The principle of the instrument's design is based on the flow of heat in metal rods as expressed by Fourier's equation. Thermally insulated rods of aluminum, copper, or silver are employed as the heat conductor between the body surface and a reference temperature mass of either water or metal. The temperature difference of the rod ends is a function of the heat flowing in the rod and is measured by a thermocouple-galvanometer arrangement calibrated to read in calories per minute. The physical factors which govern the operation of the instrument have been quantitated and will be described. When applied to equilibrium conditions in the finger, the rod calorimeter yields estimates of blood flow which agree with simultaneous estimation by the photoelectric plethysmograph and other calorimetric techniques. The instrument offers the possi-

bility of fractionating heat losses from the surface of the body on a regional basis.

Influence of protein hydrolysates on the production of nephrosclerosis and hypertension by anterior-pituitary preparations. By R. DE GRANDPRÉ (by invitation), J. L. PRADO (by invitation), P. DONTIGNY (by invitation), J. LEDUC (by invitation) and H. SELYE. *From the Institut de Médecine et de Chirurgie Expérimentales, Université de Montréal, Montréal, Canada.* Following our previous studies concerning the influence of diets on the production of hypertension and nephrosclerosis by lyophylized anterior-pituitary (LAP) overdosage, we investigated the effect of various protein hydrolysates in the food. Experiments on black-and-white ♂ rats (sensitized by unilateral nephrectomy, castration and a high NaCl diet), treated with LAP, showed that diets whose nitrogeneous constituents were 15% of casein plus an equivalent amount of protein hydrolysate were as effective in producing hypertension and nephrosclerosis as a ration containing 30% casein. "Essenamine" (a partially degraded lactalbumin), "Parenamine" (almost complete acid casein hydrolysate), "Amigen" (enzymatic casein hydrolysate) and "Bacto-peptone" added to a basic 15% casein diet were the ingredients used. In an additional group of 8 rats, the total amount of nitrogen in the diet was furnished as the hydrolysate "Amigen" and even here a high incidence of hypertension and nephrosclerosis was obtained. All the hydrolysates tried were approximately equally effective in inducing nephrosclerotic changes and hypertension.

It is tentatively concluded that the artificial hydrolysis of proteins does not destroy the factors indispensable for the production of anterior-pituitary hypertension and nephrosclerosis; hence these could be the amino-acids contained in the protein molecule. However in LAP treated rats receiving a diet which contained 15% casein and one of the following amino-acids in an amount corresponding to that contained in a 30% casein diet, nephrosclerosis did not ensue: glutamic ac., glycine, methionine, cystine, tryptophane, valine, histidine, lysine, leucine, phenylalanine, aspartic ac., isoleucine, norleucine, arginine.

Mechanism of Inactivation of α-estradiol by rat liver "in vitro". R. H. DE MEIO (by invitation), A. E. RAKOFF (by invitation), A. CANTAROW and K. E. PASCHKIS. *Jefferson Medical College, Philadelphia, Pa.* The inactivation was studied incubating either male rat liver slices (100 mg.) or homogenate (10 per cent) in 5 ml. of Krebs solution, phosphate buffer pH 7.4, without glucose for two hours at 37.5°C. Usually 1 γ of α-estradiol per milliliter was used. The amount of biologically active material present at the end was determined by bioassay in mice. Complete inactivation by liver slices was observed whether the gas phase was oxygen or air. In nitrogen or with boiled slices no inactivation took place. The 10 per cent homogenate prepared with the Waring blendor or by crushing the frozen liver in a mortar to a fine powder produced from 40 to 70 per cent inactivation. No change in the behavior of the homogenate was observed when the Fuhrman and Crismon "intracellular medium" was substituted for the Krebs solution or when boiled liver extract or glucose was added to the medium. The inactivation by liver slices inhibited by capryl alcohol, but not by cyanide, azide, thiourea, malonate, fluoride or monoiodoacetate. The addition of methylene blue to slices incubated in nitrogen brought about a 40 to 70 per cent inactivation. Nicotinamide and DPN added to the homogenate increased the inactivation by approximately 50 per cent. Our results indicate that no appreciable inactivation is due to conjugagation of the estradiol. As a consequence of our findings we may conclude that the system cytochrome-cytochrome oxidase does not participate, or if it does, only to a minor extent, and that the inactivation is probably produced by a dehydrogenating system or systems.

The spread of muscle action potentials from active to inactive areas. J. S. DENSLOW and DAVID M. GRAHAM-SERVICE (introduced by IRVIN M. KORR). *Still Memorial Research Trust, Kirksville, Missouri.* The presence, absence and degree of muscle action potentials shown in the electromyogram appear to depend on (a) the number of units (or fibers in denervated muscle) which are active and (b) the characteristics and placement of electrodes which are used. The present studies were done to determine the most effective method of measuring spatial differences between active and inactive muscle by the electromyogram. Needles enamelled to ± 1 mm. of the tip were inserted into the hind limb muscles of the decerebrate cat and paired with other enamelled or bare needles. Certain muscles were denervated and others were left intact. The following observations were made. (A) That with an enamelled electrode in an innervated muscle and one in a contiguous denervated muscle, the electrodes being less than 3 mm. apart and paired with a common reference electrode, the action potential from an active single unit or group of fibers in the innervated muscle might or might not pass the epimysium of both muscles and be recorded from the electrode in the denervated muscle. (B) When the action potential from an active unit in the innervated muscle was picked up in denervated muscle the voltage from the latter was usually extremely small. (C) When two electrodes were placed in an "electrically silent" denervated muscle and when there was massive contraction in nearby muscle, action potentials were observed which were indistinguishable in

character from those recorded from electrodes in the contracting muscles.

Liver regeneration in the rat. R. W. DENTON (by invitation) and A. C. Ivy. *Dept. of Clinical Science, Univ. of Illinois College of Medicine, Chicago.* This paper shows that liver tissue when fed to partially-hepatectomized rats facilitates the rate of regeneration of the rat's liver.

The method of the experiment was to compare the dried weights of the liver mass gained in the control (fed dog food) and the "treated" (fed pigs' liver) groups. Brues *et al* showed that the left lateral and median lobes constituted about 68.5% of the total liver mass. We removed these lobes and calculated the 31.5% remaining. The pigs' liver fed was coagulated by bringing it to a boil in order to reduce putrifacation and enhance its being eaten. We present only data on rats not losing over 15 grams of body weight in either group. No special pre- or post-operative feeding was practiced; the rats ate *ad libitum*, and they were kept in separate cages. After 10 days, the rats were sacrificed and the increment of liver obtained as the difference between the weight at necropsy and the calculated mass left in at operation. In 29 control rats, averaging 216 $\pm$ 5.6 grams body weight, the liver increment regenerated was 1.144 $\pm$ 0.03 grams, and in 22 "treated" rats averaging 218 $\pm$ 7.2 grams, the increment was 1.652 $\pm$ 0.06 grams. The difference of 0.508 grams was statistically "significant" by having a critical ratio of 7.

The dry/wet weight ratios showed the regeneration livers to have returned at least to normal consistency. No effect was obtained by injections of thiamine (200 micrograms/day) or with crude or purified antipernicious anemia liver extracts.

The stimulating effect of glycols and their polymers on the tarsal receptors of blowflies. V. G. DETHIER (by invitation) and L. E. CHADWICK. *The Johns Hopkins University and the Medical Division, Army Chemical Corps, Army Chemical Center, Maryland.* The stimulating effect of three homologous series of glycols on the tarsal receptors of blowflies has been studied by means of techniques previously used in the study of aliphatic acids and alcohols. When the glycols are compared to the corresponding alcohols it is seen that the introduction of the second hydroxy group reduces the stimulating effectiveness of the compound. Of the three series, that represented by the group ethylene, trimethylene, decamethylene is the most stimulating. Members of the series represented by ethylene glycol through polyethylene glycol 1540 show increasing stimulative efficiency as the series is ascended.

A study of the effects on the intravenous injection of hypertonic solutions on the heart rates of cats and dogs. INGRITH J. DEYRUP (by invitation) and WILLIAM W. WALCOTT. *Department of Physiology, College of Physicians and Surgeons, Columbia University.* Circulatory changes following intravenous injections of small volumes of 20% NaCl and 50% glucose have been studied in cats and dogs by means of femoral arterial pressure tracings. One of the results of such injections is profound slowing of the heart, which is absent after bilateral vagotomy. Various lines of evidence suggest that the effect depends on reflex, rather than direct stimulation of the medullary vagus center. Thus, in cats, the bradycardia was elicited consistently when 20% NaCl was injected into peripheral veins or into either the right or left auricular appendage, but was observed in only one out of six experiments in which hypertonic salt solution was injected into the ascending aorta. Analysis of the latent period of the vagal response, particularly in comparison with the longer latent period of the gasp reflex following sodium cyanide injection, suggests that the bradycardia results from stimulation of receptors located, at least in part, within the heart itself. Both the afferent and efferent neural path of the reflex are present in the vagi. Thus, the effect is not abolished by removal of both sympathetic chains from the stellate ganglia to T_7, but may be eliminated by injection by atropine sulfate.

Reflex bradycardia following the injection of hypertonic solutions was observed in decerebrate cats and dogs, and in cats but not in dogs under nembutal. It appears to be a reflex pattern related to the known cardiac adjustments resulting from stimulation of receptors in the venae cavae and in the heart itself.

The mechanics of Starling's law of the heart. JOSEPH R. DIPALMA and RICHARD A. REISS (by invitation). *Department of Physiology, Long Island College of Medicine, Brooklyn, N. Y.* Myographic measurements by two new techniques of the contractile force of the anaesthetized cat's heart were made. Simultaneous recordings of the intraventricular pressure were also obtained. With increased intraventricular pressure (clamping the aorta or pulmonary artery) the contractile force is increased. Proof was obtained in the beating heart that cardiac muscle resembles skeletal muscle in regard to the phenomenon of initial tension. This explains the fact that the contractile force of the heart muscle increases with a rise in intraventricular tension. On the other hand, the contractile force decreases with increased venous return, and the opposite occurs with decreased venous return. These latter findings are seemingly contradictory to the usual statement of Starling's Law of the Heart. However, they do not contradict the original conception of the law as related to energy production. The findings do indicate that the heart during systole follows the physical laws of an elastic sphere such as a soap bubble in which the contractile stress increases as the radius decreases. Thus the force of

contraction of the heart muscle depends merely upon its initial length (diastolic size) not upon the initial stretch of muscle causing a greater subsequent contraction. Because of this fact the analogy which in the past has been drawn between the phenomenon of initial length of cardiac muscle as compared to that of skeletal muscle is not tenable.

Observations on the peripheral circulation in neurogenic hypertension. ISABELLE DOUGHERTY and SISTER M. A. C. DAY (introduced by A. B. HERTZMAN). *Department of Physiology, St. Louis University, School of Medicine.* Changes in the peripheral circulation of dogs during neurogenic hypertension have been studied. Photoelectric plethysmograms are recorded from various areas and pressure pulses are optically recorded from the femoral artery. The experiments are acute, the recording being continuous throughout the control period, the period of operation (removal of the buffer and sympathetic nerves, after Heymans) and the subsequent hypertensive period. The volume pulse amplititude can be correlated with the variations in pulse pressure. The blood pressure usually shows an immediate rise after the denervation, a rise to about 240/160 mm. Hg being obtained within 2 to 5 minutes. In some dogs the rise in blood pressure after the denervation is soon followed by a marked increase in volume pulses in the intestine and muscle, decreased blood pressure and death. In other dogs the hypertension develops more rapidly, rises to higher levels, and persists for hours. In such cases decreased volume pulses are usually seen in the skin, pad, intestine, and muscle. The anesthetic used influences the height of the rise in blood pressure.

The basal metabolic rate of Georgia medical students. PHILIP DOW and ROBERT H. SHULER (by invitation). *Dept. of Physiology, Univ. of Georgia School of Medicine, Augusta, Georgia.* Records have been kept of the basal metabolic rate determinations of the last ten sophomore medical classes (over 500 individuals). Procedures were carried out by the students on each other; techniques were supervised by the physiology teaching staff, and all records and calculations have been checked by the authors. A period of eight years is covered, including classes in all four seasons of the year.

The overall average runs between twelve and thirteen percent below the 1936 Mayo standards. This is lower than most reports from the South, but about the same as that recorded by Eaton in 1939 for a smaller group of similar age in New Orleans. Figures for age, sex, and seasonal differences are of questionable significance. Further statistics are in process of compilation. Limitations and implications of the findings will be discussed.

The authors wish particularly to emphasize the relevance of these results to the necessity for the use of local controls in forming diagnostic conclusions. This caution has appeared repeatedly in the literature on metabolism, but unfortunately not in the instructions and tables furnished with clinical apparatus.

Effect of convulsant agents on partially isolated neurones of the central nervous system. C. G. DRAKE (by invitation), and G. W. STAVRAKY. *Department of Physiology, Faculty of Medicine, University of Western Ontario, London, Canada.* It is generally believed that the highest levels of the central nervous system are most sensitive to convulsants such as thujone, camphor and metrazol, and that decortication reduces the susceptibility of animals to convulsions. This concept is contrary to the "Law of Denervation" according to which isolated neurones should become more sensitive to chemical stimulating agents. With this in view the relative sensitivity of different regions of the central nervous system to various convulsant drugs was reinvestigated systematically in 37 sacrifice experiments on cats after aseptic semisection of the spinal cord or after semidecerebration carried out 4 days to 3 months previously, and in 28 chronic cats with removal of one motor cortex, a frontal lobe or a cerebral hemisphere. It was found that in both sets of experiments decentralized neurones reacted to smaller quantities of convulsant agents than intact ones, and that camphor, metrazol and picrotoxin evoked greater and more prolonged responses from decentralized neurones when sufficient time was allowed for sensitization to take place. This was ascertained on tracings of muscular contractions in sacrifice experiments and in moving camera recordings of convulsions induced in chronic animals. Chemically induced convulsions play a prominent part in the study of epilepsy and in the treatment of some mental derangements and it is felt that this investigation may contribute to the understanding of the mechanism of action of convulsant agents on the nervous system in which pathological processes are present.

Acute effects upon the lungs of dogs of large intravenous doses of alpha-naphthyl thiourea (ANTU). CECIL K. DRINKER and ESTHER HARDENBERGH (by invitation). *Dept. of Physiology, Harvard School of Public Health.* Dogs under nembutal anesthesia and given intravenously 2 per cent alpha-naphthyl thiourea (ANTU) in propylene glycol, 1 cc. per kilogram, develop fatal pulmonary edema. The first evidence of changes leading to edema is increase in the flow of lung lymph. Greater rate and minute volume of breathing follow. Blood gas analyses indicate that the hyperpnoea of rapidly developing pulmonary edema is peripheral in origin. Detailed protocols are given for two typical experiments.

The effects of burns on kidney function. ARTHUR J. DZIEMIAN (introduced by W. FLEISCHMANN). *Physiology Section, Medical Division, Army Chem-*

ical Center, Maryland. Goats with severe third degree flame burns, covering about fifty percent of the body surface area, were studied. Kidney function, including glomerular filtration rate, effective renal plasma flow, filtration fraction and maximal tubular reabsorptive capacity, was determined in the normal animal, the burned animal, and burned goats treated with plasma or whole blood. Most severely affected was the effective renal plasma flow, which decreased in all burned animals. There were two classes of effects in regard to glomerular filtration rate and maximal tubular reabsorption after burning. In one group they decreased continuously until death. In the other group there was an initial sharp decrease in the filtration rate and reabsorptive capacity, followed by a return to normal or near normal values. Preterminally these latter values fell precipitously. Neither blood nor plasma infusions had any apparent effect on kidney function in the burned goat. The greater decrease in plasma flow as compared with filtration rate, with an increased filtration fraction, indicated a constriction of the efferent arterioles of the kidney.

The influence of hypotension on coronary blood flow, cardiac oxygen metabolism and cardiac work. J. E. ECKENHOFF, J. H. HAFKENSCHIEL, E. L. FOLTZ and R. L. DRIVER (introduced by CARL F. SCHMIDT). *Department of Pharmacology and Harrison Department of Surgical Research, School of Medicine, University of Pennsylvania.* The effect produced by hypotension upon coronary blood flow and cardiac oxygen metabolism has been measured in 8 intact dogs, lightly anesthetized with pentobarbital sodium. Coronary blood flow was estimated by the N_2O method, venous blood being collected by a catheter in the coronary sinus (Am. J. Med. Sci., December 1947). Cardiac output was calculated by the Fick principle. Hypotension was produced by the intraspinal injection of procaine hydrochloride in 6 instances and by the intravenous injection of Etamon in 2 cases.

Blood pressure and cardiac output declined in all experiments. Coronary blood flow was essentially unchanged in three experiments in spite of considerable reductions in blood pressure and cardiac output. In three other dogs, coronary flow was reduced markedly; in these three instances, however, blood pressure and cardiac output were correspondingly markedly reduced. In the eight experiments blood pressure was decreased an average of 29% and cardiac output an average of 42%. These two factors were responsible for a 55% reduction in left ventricular work. Since coronary flow was diminished an average of 25%, it follows that coronary blood flow remained relatively excessive for the amount of work that the heart was performing in hypotension. The mechanical efficiency of the heart declined an average of 36%, but this does not mean that the heart was less able to perform the work demanded of it.

A method for the study of gastric secretion in vitro. LESLIE E. EDWARDS and CAROLYN TROWBRIDGE EDWARDS (introduced by R. W. RAMSEY). *Department of Physiology and Pharmacology, Medical College of Virginia, Richmond.* An apparatus is described by means of which secretion of the isolated gastric mucosa can be studied in vitro. The method is, in principle, similar to that used by Gray (A. J. P. **130:** 327, 1940) to study secretion by frog mucosa in that a piece of the mucosa, stripped away from the remainder of the stomach, is clamped between two separate chambers in such a way that the submucosa side is bathed by a nutrient medium containing any desired substrates, drugs, etc. In the present apparatus, the oxygen tension can be closely regulated to meet the requirements of the tissue and the true, unmodified secretion can be collected from the other chamber. The whole is mounted on a shaker in a water-bath at 37.5°C., making it suitable for study of secretion by warm-blooded animals. Under these conditions, the mucosa will live for at least three hours, as demonstrated by normal oxygen uptake from a Warburg flask at the completion of the experimental period. Secretion of both acid and pepsin has been obtained from the mucosae of rat and rabbit by this method. The apparatus is well adapted for study of the conditions necessary for gastric secretion and of the action of various stimulating and inhibiting agents directly upon the secreting cells.

Factors influencing in vitro secretion of pepsin. CAROLYN TROWBRIDGE EDWARDS and LESLIE E. EDWARDS (introduced by R. W. RAMSEY). *Department of Physiology and Pharmacology, Medical College of Virginia, Richmond.* Gastric secretion from the isolated gastric mucosa was collected in vitro by the method described in the preceding communication. Studies were made, using the rat and the rabbit, on the effect of variations of the composition of the nutrient medium (protein, amino acids, etc.) on the production of pepsin. Further studies were made on the stimulating or inhibiting action of common autonomic drugs. Experiments indicate that nitrogen-containing compounds are necessary for the production of pepsin and that acetylcholine with eserine (and, to a lesser extent, eserine alone) stimulates its secretion in vitro.

Further observations on intravascular temperature in man. LEON EISENBERG (by invitation) and H. C. BAZETT. *Physiology Department, University of Pennsylvania.* Any rigorous attempt to distinguish reflex from central mechanisms governing temperature regulation presupposes a knowledge of "central" temperature—in this case, the temperature of the blood perfusing the hypothalamus. Analyses in which rectal temperature is regarded as "central" temperature can be correct only (a) if temperatures of rectum and hypothalamus are identical or (b) if these temperatures show identi

cal direction and rate of change under the conditions investigated. The experiments herein reported, employing internal jugular vein or common iliac artery temperature as a first approximation to that of the hypothalamus, fail to substantiate completely either of these assumptions so it suggested that although qualitative generalizations are unlikely to require modification, quantitative conclusions heretofore obtained must be reassessed.

In this study, fine wire (38–42 gauge) copper-constantan thermocouples enclosed in polyvinyl plastic catheters have been introduced for distances of 0.5–20 cm. within the lumen of femoral, brachial, radial, and dorsalis pedis arteries, internal jugular, femoral, antecubital, wrist, and ankle veins, and have been left in place for experimental periods of up to 6 hours so that simultaneous determination of as many as 6 intravascular as well as rectal and cutaneous temperatures have been possible. Whereas the more central vascular and the rectal temperature do not differ remarkably under steady state conditions, during periods of environmental change these values deviate significantly from each other, show different time characteristics of their rate of change, and may even, though rarely, change in opposite directions.

Effects of positive intrapulmonic pressure on muscular contraction. LEONARD H. ELWELL (by invitation) and JOHN W. BEAN. *Physiology Laboratory, University of Michigan, Ann Arbor.* A continuation of an earlier study by one of us on the effects of increased intrapulmonic pressure on muscular contraction showed that such pressure (260 mm. H_2O for 3 to 5 minute periods) commonly enhanced the response of the Tibialis Anticus Muscle to intermittent single shock stimulation (3 second intervals) applied to the intact or the peripheral end of the sectioned peroneal nerve in anesthetized or decerebrate dogs.

The possibility that this enhancement, gradual in onset and persisting throughout the period of increased pressure, might have been due either to an initial temporary increase in alveolar O_2 tension, or to the subsequent apnea, induced by the pressure was eliminated by appropriate adjustment of the O_2 tension of the compressional air, and by the use of constant artificial ventilation, respectively. The volume flow of blood through the leg was markedly diminished and the pH of the arterial blood measured continuously by glass electrode was decreased. Moderately severe hemorrhage which decreases volume flow of blood induced similar enhancement. It is tentatively concluded that the enhancement resulted from an alteration in the removal of Metabolites consequent upon the decreased blood flow. and since this enhancement has been missing or less pronounced in direct stimulation of the curarized muscle, it is inferred that the neuromyal function is primarily involved. Sympathectomy in an earlier study did not eliminate the response to pressure, but the possible involvement of a circulating hormone as adrenalin has not been ruled out.

The effect of Aminophyllin on cardiac output and renal hemodynamics in man. DORIS J. W. ESCHER (by invitation), RAYMOND E. WESTON, GEORGE LEINER (by invitation), LOUIS LEITER (by invitation), and SYBIL GOLDAT (by invitation). *Medical Division, Montefiore Hospital, New York 67, N. Y.* Chasis *et al* (J. Clin. Invest. **17**: 683, 1947) observed that in normal men Aminophyllin (Theophylline ethylene diamine) produces an immediate, marked, sustained increase in GFR with a sustained decrease in RPF, after an initial brief period during which RPF also is increased somewhat. Our group has found that in patients with congestive failure Aminophyllin produces a similar immediate increase in GFR with an immediate, but smaller, increase in RPF, which is sustained, unlike that observed initially in normal subjects. To analyze the cardiovascular-renal actions of Aminophyllin, GFR, RPF, and/or cardiac output (by the direct Fick Principle) were determined in human subjects before and after the rapid intravenous administration of Aminophyllin (0.72 grams).

In normal subjects, Aminophyllin generally produces a significant increase in cardiac output, which persists ten to twenty minutes and parallels the brief period of increase in both RPF and GFR, described by Chasis *et al*. The cardiac output, then, returns to normal but the GFR remains elevated for as long as an hour, despite the fall in the RPF to below control levels after the initial renal hyperemia. In chronic congestive failure, as McMichael (Clin. Sci. **6**: 125, 1947) has reported, Aminophyllin produces a marked increase in cardiac output, which is sustained for as long as an hour and probably explains the persistent renal hyperemia observed in these cases. The sustained increase in GFR and filtration fraction observed in both normals and cardiacs probably results, in part, from a direct renal action of Aminophyllin.

The effect of nicotine on maze learning ability of albino rats. J. M. ESSENBERG (introduced by GEORGE CLARK). *Department of Anatomy, The Chicago Medical School, Chicago 12, Ill.* Eighteen young, sexually mature albino rats were selected for the experiment. They were males and females from four litters and were treated as such throughout the experiment. Food was withheld from the animals 12 to 18 hours before they were placed in the maze. The Lashley general type of maze was adopted for this work. The first part of the experiment consisted in learning of the maze. The animals were considered ready for nicotine when they could make ten error-less runs for at least two consecutive days. This was followed by daily injections of

one-half cubic centimeter nicotine-saline solution of 1:1000 or 1:2000, subcutaneously and in some instances intraperitonially. The maze running began in some instances soon after recovery from the injection, but in most cases, 24 hours later. A record of behavior but particularly the time required and the errors made for 10 runs were kept for each rat. The experiment was designed for 18 to 24 months. Some of the rats were eliminated from the experiment because of lack of intelligence or because of illness. Controls received similar treatment except the nicotine. Of all the animals treated with nicotine, there was a gradual increase in the number of errors per ten runs. As to the time required, a few showed slight differences, but the majority ran high scores, two or three times the control limit.

Effects of cold on infant rats. JANET FAIRFIELD (introduced by E. F. ADOLPH). *Dept. of Physiology, The University of Rochester, Rochester, N. Y.* Rats from 0 to 17 days of age were cooled in an atmosphere of oxygen of 2°, 5°, 10°, or 20°C. within a glass respirometer surrounded by water. When more than 3 days old, but not before, they manifested large increases in oxygen consumption, which soon diminished as the body temperature fell. In all individuals less than 11 days old, exposed to 3° or lower, the consumption reached zero; it could remain there for 1.7 hours and still be followed by complete recovery of the animal and acceptance by the mother rat. The rate of heart beat as observed by electrocardiograms slowed, and the beats disappeared at body temperatures between 9° and 4°. These temperatures are below those at which the mature rat's heart stops. When rewarmed in surroundings of 35°, the heart beats, breathing, limb movements and oxygen consumption reappeared within a few minutes. Asystole lasting 1.3 hours was followed by complete recovery. After two hours of cold exposure, ten percent of the animals failed to recover; their hearts beat irregularly with atypical QRS waves which suggested injury, possibly from anoxia, to the cardiac conduction system. After two hours of exposure all hearts resumed beating during rewarming; subsequent failures of beat occurred only in rats whose body temperatures had been below 8°. Majority survival at 3° body temperature contrasts with adult rats which die at 14° to 16° body temperature. The contrast classes newborn rats as poikilothermic within brief periods of cooling.

On the mechanism of the action of di-isopropyl fluorophosphate. EMILY A. FELD (by invitation), MORTIMER A. ROTHENBERG (by invitation) and DAVID NACHMANSOHN. *Department of Neurology, College of Physicians & Surgeons, Columbia University, New York.* The irreversible inhibition of cholinesterase by di-isopropyl fluorophosphate (DFP) requires a measurable period of time dependent upon temperature, DFP concentration, condition of the enzyme, etc. This feature made it possible to establish a close parallelism in all cases between the rate of irreversible enzyme inactivation and irreversible abolition of conduction. Whatever factor is changed, the two events are influenced in the same way. It has not been possible under any condition to dissociate conduction and cholinesterase activity. Claims to the opposite Boyarsky, Tobias & Gerard) are based on the use of inadequate techniques. Under the experimental conditions used but determining the enzyme activity with the proper enzyme to substrate ratio, presence of cholinesterase has been demonstrated (Feld, Grundfest, Rotherberg, Nachmansohn, in press). Death of animals following DFP injection coincides with the disappearance of cholinesterase. In surviving animals, cholinesterase is always present, although in a concentration which is on the average about 25 per cent of the initial. The question of enzyme excess and some other aspects of DFP poisoning will be discussed. All findings support the assumption that the toxicity of the compound has to be attributed exclusively to the inactivation of cholinesterase, the enzyme without which the vital process of conduction is impossible.

Decrease in the radiosodium and thiocyanate spaces during growth. FRANCIS X. FELLERS (by invitation), and HENRY L. BARNETT (by invitation), HELEN MCNAMARA (by invitation) and KENDRICK HARE. *The New York Hospital and Department of Pediatrics, Cornell University Medical College, New York City.* Measured amounts of sodium thiocyanate and $Na^{24}Cl$ were injected intravenously into 40 subjects including premature and full term infants, children and adults. Analyses of serum and urine collected from 1–12 hours later permitted the calculation of the volume of fluid in which these substances were distributed. The results are summarized in the table, in which the available space is expressed in per cent of body weight. Serial observations were made on 2 babies over a period of 5 months, showing a decrease in the moiety of body weight available as a solvent for thiocyanate and sodium. Simultaneous determinations of thiocyanate, Na^{24}, mannitol and inulin spaces indicate that the latter two substances are unsatisfactory for comparison since their spaces never attained a constant value but continued to increase throughout the period of observation.

Weight	No. subjects	Thiocyanate space (2½–4 hour)	Radiosodium space (2½–4 hour)
kg.		%	%
2.1– 4.5	12	37.7	42.0
4.6–10.0	12	31.7	34.6
10.1–40	11	29.0	33.4
60 –75	5	20.7	26.4

Studies on the stability, inhibition and activation of fibrinolytic protease (tryptase, plasmin). JOHN H. FERGUSON, and (by invitation) JESSICA H. LEWIS, P. W. BOYLES, B. L. TRAVIS, and E. B. GERHEIM. *Dept. of Physiology, Univ. of North Carolina, Chapel Hill.* Employing fibrinolytic and fibrinogenolytic assay methods, data on the relative activity of plasma protease preparations of human, bovine, and canine origin are presented, together with a study of the stability of these materials in dry lyophilized state and in solutions stored at −20°, 0°, 28° and 37°C. Attempts were made to stabilize the activity of these preparations at room temperature using various materials, e.g. acacia, crystalline soybean trypsin-inhibitor (SBI), etc. Mixtures of protease and SBI reach optimal fibrinolytic activity at a time much later than that at which untreated enzyme has lost most of its potency. Stability and activity of SBI at various dilutions and the effects of temperature of storage are followed.

The action of SBI is compared with the antiproteolytic activity of natural serum and of the bovine "antifibrinolysin" of E. C. Loomis. Additional experiments on the stability of streptokinase (enzyme-activator) are described, together with some data on the stability and interactions of the enzyme precursor.

The extension of these studies to preliminary investigations of *anti*enzyme and *pro*enzyme in normal and pathological sera will be noted.

The physiological basis for the internal ventilation of clothing. E. S. FETCHER, S. I. RAPAPORT (by invitation) and JOHN F. HALL (by invitation). *The Aero Medical Laboratory, Wright Field, Dayton, Ohio.* Neither physiological regulation nor insulative clothing can protect man exposed to a wide range of rapidly changing temperatures, or to very high temperatures. These conditions are of common occurrence in aviation, industry, and the armed services. This problem can be met by replacing the ambient environment by an individual environment created by blowing air under clothing and near the skin. This personal environment can be kept constant over a wide range of ambient temperatures by regulation of ventilating air temperature and flow to compensate for heat leakage through the outer clothing.

Subjects wearing a ventilating harness, and clothing designed for comfort at about +50°F., Have been exposed to −30, 0, +70, +120, and +180°F. At −30 and +180°F. about 55 CFM (4.1 lbs./min.) of 120 and 60°F. air respectively, were sufficient to maintain thermal equilibrium, and hence comfort, indefinitely. Under proper conditions, the hands and feet require no artificial heating or cooling (see Rapaport, Fetcher, and Hall, this issue). The general relationships between ventilating air temperature and flow, and metabolic rate, clothing, and environment are defined; the physical and physiological requirements are delineated.

The influence of substances affecting body temperature on oxygen consumption and glycolysis in brain. J. FIELD, C. N. PEISS (by invitation) and V. E. HALL. *Department of Physiology, Stanford University, California.* Previous work in this laboratory suggested the hypothesis that when the metabolic rate of the temperature regulating center is increased the center behaves as though its temperature were raised and regulates body temperature at a subnormal level. It thus becomes of interest to examine the influence of substances which raise or lower body temperature, possibly by altering the level at which temperature is regulated (see abstract of Hall, Grant and Field) upon the metabolism of the center. Direct investigation of the metabolism of the center itself has not yet been feasible. However, the available evidence indicates that the course of carbohydrate catabolism is qualitatively similar in the several parts of the central nervous system. Accordingly we have investigated the effect of substances modifying body temperature on the oxidative and glycolytic metabolism of rat cerebral cortex slices which may possibly reflect the metabolic picture in the hypothalamic centers. The influence of graded concentrations of magnesium (as the chloride), typhoid-paratyphoid vaccine (T.P.T.), 2,4-dinitrophenol (DNP) and antipyrine on oxygen consumption and anaerobic glycolysis in rat cerebral cortex at 37.5°C. was studied. Conventional manometric methods were used. From this evidence it appears that neither of these over-all metabolic processes is directly related to possible change in the set level of the temperature regulating center.

Substance	Concentration range	Metabolic effect		Effect on body temperature
		Q_{O_2}	$Q_A^{N_2}$	
Mg *mM/l*.....	0.15–5.2	0	+	−
T.P.T.....	1/50000–1/2000	0	0	+
DNP *mM/l*.....	0.11–13.4	+ to −	0	+
Antipyrine *mM/l*....	0.054–5.4	0	0	−

0 denotes mean values same as control, + above control, − below.

Urinary tract infections and antibiotics. EDMUND P. FINCH (by invitation) and SEWARD E. OWEN. *Veterans Administration, Hines, Illinois.* Urines from two hundred and seventy paraplegic patients with urinary tract infections were studied. Bacterial organisms of thirty seven different species were isolated in a series of nearly two thousand culture classifications. Early cases displayed an average of two different organisms while later each case returned an average of five different organism species. The increase is undoubtedly due to the impossibility of sterilizing body parts and to the necessity

for repeated catheterization due to incomplete drainage and incompetent sphincters. Seventy five per cent of the organism types found were gram negative. Aerogenes occurred in 62.4% of all cases, proteus in 52.8, pseudomonas in 43.4, E. coli in 40.5, enterococci in 39.8, paracolon in 33.2, achromobacter in 31.4, streptococci in 30.7, staphylococci in 24.1, micrococci in 12.4, alcaligenes in 9.1, diphtheroids in 8.4, yeasts in 1.8 and three other types in less than 1% of all cases. Separate colonies of the same type organism from the same plate often displayed considerable variation in sensitiveness to penicillin or streptomycin. A more exact prediction of organ sensitiveness to the antibiotics and their probable efficiency in treatment can only be obtained by combining several typical colonies of the same organism and determining the antibiotic resistence of this mixture.

Effect of magnesium on alkaline phosphatase as influenced by pH, enzyme concentration and aging. CLARY J. FISCHER (by invitation) and ROY O. GREEP. *Harvard School of Dental Medicine.* Boston, Massachusetts. Purified alkaline phosphatase standing at room temperature loses its activity more rapidly at pH 9.5 than at pH 6.5. The activity can be partly or completely restored when an optimal concentration of magnesium ions is added to the substrate during activity determination. On standing with magnesium phosphatase loses its activity more rapidly regardless of pH and can no longer be activated by adding magnesium to the substrate. Adding magnesium to the substrate during incubation of fresh (highly active) phosphatase produces much less activation than similar treatment of aged samples having reduced activity. This difference can be explained by the assumption that the fresh sample contains a sufficient amount of metal for its complete activation. The activating effect produced by adding an optimal concentration of magnesium to the substrate steadily increases with the duration of standing at pH 6.5 whereas on standing at pH 9.5 there is an initial decline suggesting participation by the recovered inorganic group of phosphatase at this pH. These observations suggest that phosphatase is a metal protein and that the inactivating and reactivating effects involve the specific nature of the metal group of the enzyme protein.

Changes in zymohexase activity during denervation atrophy of skeletal muscle and their retardation by appropriate electrical treatment. ERNST FISCHER and RUSSEL V. BOWERS (by invitation). *Baruch Center of Physical Medicine, Medical College of Virginia, Richmond.* Myogen was extracted from mash of normal and denervated rabbit gastrognemii and purified by dialysis. The zymohexase activity of the crude extract and the purified preparation was determined by the method of Herbert et al. For normal muscle no enzyme activity was lost by dialysis, while for denervated muscle a loss up to 10% was observed. The amount of protein extractable per gm muscle increases slightly until the second week after denervation, but the enzymatic activity per mg protein declines slowly during the same time. Since the decline of the specific activity nearly balances the increase in extractable protein, and since the small remaining decline in total enzymatic activity calculated per gm muscle can be explained by the relative increase of connective tissue, it is concluded that zymohexase activity is not altered during the first two weeks of denervation. From the third week on, the extractable protein as well as its specific activity diminishes rapidly.

Appropriate electrical treatment has no beneficial effect upon the decline in extractable protein, but diminishes considerably the decline in zymohexase activity per mg protein. In consequence, the total loss in zymohexase activity per gm muscle is about halved by the electrical treatment. Some of the experimental findings indicate that the changes in zymohexase activity are of secondary nature and are rather independent of the wasting of the muscle.

The lipids of the rat brain in choline deficiency. PIERO P. FOA and HARRIET R. WEINSTEIN (by invitation). *Department of Physiology and Pharmacology, The Chicago Medical School, 710 S. Wolcott Avenue, Chicago, Illinois.* It has been shown that the lipid composition of the brain is not modified by choline added to an adequate stock diet nor by the amount or the nature of the dietary fat. The only exception to this rule appears to be a slight and temporary decrease in brain phospholipids after protracted cholesterol feeding. (References in A. V. Stoesser et al., Proc. Soc. exp. Biol. and Med., **32**: 761, 1935). To our knowledge the brain lipids of animals fed a choline deficient diet have not been studied. The following experiment was, therefore, made. Forty male hooded rats (73 to 215 g.) were paired and given the following diet; lard 20, corn oil 20, casein 15, sucrose 20, starch 18, cod liver oil 3, salts 4, plus 1 yeast tablet per day, for 18 to 25 days. One rat in each pair received a diet supplemented with 0.2% choline hydrochloride. The rats were killed by stunning. Brain and liver were immediately removed for analysis. All lipids were determined according to M. H. Hack (J. Biol. Chem. **169**: 137, 1947) with the exception of cholesterol which was determined according to W. R. Bloor (J. Biol. Chem. **24**: 227, 1916). The lipid composition of the liver was used as a criterion for choline deficiency. No significant differences were found in the total lipid, total phospholipid, sphingomyelin, acetylphosphatides and cephalin, and cholesterol content of the brain of the choline deficient rats and the pair-fed animals receiving choline.

Coagulation in plain and partially heparinized

plasma. R. H. K. FOSTER. *Department of Pharmacology, St. Louis University School of Medicine.* The degree of coagulation was visually estimated (J. Amer. Pharm. Assoc. **36:** 243, 1947) on recalcified heparinized or unheparinized citrated beef or sheep plasma (stored in frozen state). The clot was broken up, washed with saline and centrifuged in cellulose nitrate tubes at 18,000 G. After compaction into a rubbery deposit it was washed with water, dried and weighed. Unheparinized plasma gave clots weighing 4 to 10 mg./ml. of plasma. Other serum constituents besides fibrin were undoubtedly represented in the weight. Centrifugation during coagulation resulted in clots weighing 20% less. Clots from unheparinized plasma formed in glass, silicone-coated glass or cellulose nitrate tubes were of equal weight. At least 50% more heparin was needed to give inhibition in glass tubes comparable to that in the latter two which were about equal in effect. Clots from heparinized plasma weighed less and in approximate proportion to the degree of coagulation. In the "4+" clotting zone the clot weight decreased very slightly with increasing heparin. This was in contrast to the increase in optical density in this region. At the point where active visible inhibition commenced both the clot weight and the optical density decreased rapidly. Curves of these observations could be approximately but not exactly correlated with the curve based on degree of coagulation because of inherent errors in all three, viz: extraneous material in the weighed clot, lack of uniformity of clotting affecting the densimetric measurements and personal errors in judging the degree of coagulation.

Renal and circulatory factors in congestive failure of the circulation. D. M. FOWELL (by invitation), A. P. BRIGGS, N. C. WHEELER (by invitation), J. A. WINSLOW, JR. (by invitation), J. W. REMINGTON, and W. F. HAMILTON. *Departments of Physiology, Biochemistry, and Medicine, University of Georgia School of Medicine, Augusta, Georgia.* A series of patients in various stages of the development and remission of congestive failure is being studied. The following determinations are being made: cardiac output, intracardiac and intra-arterial pressures, peripheral resistance, circulation times, vital capacity, blood volume, thiocyanate space, glomerular filtration and sodium clearance. Response to bed rest, digitalis, theophylline-isopropanolamine (Theopropanol, National Drug Co.) and mercurial diuretics are being investigated. With clinical improvement there is, as has been observed by others, a reduction of the thiocyanate space and blood volume, an increase in vital capacity, and in the glomerular filtration and sodium clearance. Correlation of these favorable renal and clinical findings with improvement in the circulatory variables studied has not been clearly established as yet. Theopropanol produces a decided, but evanescent, increase in cardiac output with small change in the peripheral resistance. Our subjects include patients with renal impairment, and results so far show that the improvement in sodium clearance and the elimination of edema fluid are not associated with any absolute level of glomerualr filtration, i.e., not with the quantity of sodium presented to the tubules.

Respiratory dead space. WARD S. FOWLER (introduced by Julius H. Comroe, Jr.). *Department of Physiology and Pharmacology, Graduate School of Medicine, University of Pennsylvania, Philadelphia.* Respiratory dead space was measured by simultaneous and continuous measurement of volume flow and N_2 content (Lilly-Hervey nitrogen meter) of gas expired following inhalation of 99.6% O_2. The average dead space volume of 30 normal males at rest was 155 cc. (19% coefficient of variation); the average expired volume required to wash out the dead space was 317 cc. The dead space/tidal volume fraction varies in different individuals. Physiological measurements of dead space are affected by: 1) *Anatomical volume of the bronchial tree.* Maximal variations of inspiratory lung volume changed dead space by 100–150 cc. Voluntary hyperventilation and exercise hyperpnea increased dead space equally (50–100 cc.). Increase in tidal volume decreased the dead space/tidal volume fraction. 2) *Gas diffusion between terminal bronchioles and alveolar spaces.* Prolongation of inspiratory time by 1–2 seconds significantly reduced dead space volume, and breathholding during inspiration (20 sec.) caused reductions of 45–90 cc. 3) *Uniformity of gas mixing throughout the lung.* When uneven gas mixing is present, the usual methods of dead space measurement give artificially large results; this method shows directly the degree of uniformity in mixing.

Metabolic balances in the cold environment: II. Energy exchanges. JOHN A. FRANTZ and JAMES L. A. ROTH (introduced by J. W. Heim) *Physiology Branch, Aero Medical Laboratory, Wright Field, Dayton, Ohio.* Energy exchange data were obtained on human subjects residing continuously for nine days in the cold environment, −32°C., to appraise the role of a USAF emergency ration in a survival situation. Fasting in the same environment for a period of six days provided control data. Clothing assemblies and sleeping bags were ade-

	Emergency ration	Fasting
Caloric intake	1890	0
Caloric output	2843	2800
Water intake	899	875
Weight loss, gms.	307	891
Body water	178	499
Body tissue	121	389

quate to maintain normal body temperature and comfortable skin temperatures without the necessity for fatiguing exercise during fasting or subsistence upon the ration. Average daily values are expressed in terms of the standard 70 kg. man.

Progressive limitation of voluntary activity was noted in the fasting subjects. The R.Q. approached 0.72 with continued fasting, especially for activities requiring more rapid caloric expenditure. The metabolic cost for a standard work load on the treadmill was increased during low temperature exposure. This was not observed during the activities in which the hobbling effect of the clothing did not participate, *i.e.*, lying, sitting and standing quietly. Considering ability to perform psychomotor tasks, gross work capacity, and maintenance of body comfort in the fasting and non-fasting subjects, food appears to be of secondary importance to the adequacy of clothing and sleeping bag for survival in low ambient temperatures.

Correlation between signs of toxicity and cholinesterase level of brain and blood during recovery from di-isopropyl fluorophosphate (DFP) poisoning. A. M. FREEDMAN, (by invitation) and H. E. HIMWICH. *Army Chemical Center, Medical Division, Edgewood, Maryland.* Male rats were injected with Di-Isopropyl Fluorophosphate (DFP) (2.0 mg./kgm. subcutaneously) and sacrificed at intervals varying from one hour to 14 days, at which time the rats were asymptomatic. Prior to sacrifice, signs of toxicity were carefully noted, then brain, plasma, and erythrocyte cholinesterase activity was determined. Plasma cholinesterase rapidly returns to normal in a few days. Erythrocyte cholinesterase remains at zero for about 48 hours and then rapidly regenerates. Brain cholinesterase increases more than three-fold in the first 24 hours and thereafter the rate of regeneration decreases. The clinical signs of DFP toxicity diminish as the brain cholinesterase rises. The level of brain cholinesterase corresponding to the various degrees of severity of clinical signs in this recovery period is in general similar to that which which has recently been demonstrated for the acute phase.

These data suggest again that the severity of the signs are related to the brain cholinesterase activity. The correlation between cholinesterase level of brain and blood that has been demonstrated immediately after DFP injection is not found here. Further, an explanation is afforded to the findings of Harvey et al. that although there is a correlation in the human between red blood cell cholinesterase and symptoms immediately following DFP injection, when the drug is withdrawn symptoms disappear while the RBC cholinesterase remains low. This phenomenon appears to be due to the very rapid rate of regeneration of brain cholinesterase as compared to RBC cholinesterase in the first day or two after the injection.

The effect of size, sex and pregnancy on the lethality of di-isopropyl fluorophosphate (DFP). A. M. FREEDMAN, (by invitation) and H. E. HIMWICH, *Army Chemical Center, Medical Division, Edgewood, Maryland.* Previous work has revealed that newborn rats are much more sensitive to a dose of 2.0 mg./kgm. of Di-Isopropyl Fluorophosphate (DFP) subcutaneously than adult rats of both sexes weighing between 100–200 gm. In the course of experiments, it was observed that pregnant females were also particularly susceptible to the toxic effects of DFP. In order to study the importance of size and sex in the sensitivity of the pregnant female, the following preliminary mortalities were found in various groups of rats:

Rats	No.	Died	% Died
Males (100–200 gm.)	20	6	30
Females (100–200 gm.)	20	3	15
Males (250–400 gm.)	20	13	65
Females (250–400 gm.)	10	6	60
Pregnant females (200–400 gm.)	18	17	94

Thus, there is no significant statistical difference between normal males and females of the same weight. However, there is a significant difference in the susceptibility between light and heavy rats and between the latter and pregnant females of similar weight. The mechanism of this variation in susceptibility is being investigated.

Conduction of painful impulses from the extremities via the sympathetic nervous system. L. W. FREEMAN, H. B. SHUMACKER, JR. (by invitation), E. E. WAYSON (by invitation) and N. M. STAHL (by invitation). *The Department of Surgery, Yale University School of Medicine, New Haven, Connecticut.* Sympathetic denervation of the extremities often provides relief of pain in patients suffering from various vascular disorders and regularly in patients with major causalgia. When pain associated with ischemia is thus relieved in limbs, the circulation of which is improved by the procedure, one can explain the relief by section of efferent sympathetic fibers. When pain is relieved in such cases without improvement in circulation and in cases of causalgia, it is difficult to understand the mechanism on any basis other than interruption of afferent pain fibers.

Studies on dogs utilizing various responses as evidence of pain suggest that there are multiple pathways by which painful impulses arising in the lower extremities may reach consciousness. Section of the spinal cord at sufficiently high levels, anterior and posterior rhizotomy of sufficient extent, and complete bilateral sympathectomy appear to eliminate responses to acute vascular injuries. Utilizing the local vasoconstrictor response to

acute chemical injury of the femoral vein as an index of pain, experiments would indicate that afferent pain fibers are present in the sympathetic nerves, interruption of which serves as a satisfactory explanation for relief of pain.

A comparative study of the effects of several anticholinesterases on central nervous integration. JEANE SISKEL FREY (by invitation) and ROBERT GESELL. *Physiology Laboratory, University of Michigan, Ann Arbor.* All anticholinesterases studied produced coordinated hyperpnea. Carbon dioxide was most dependable, next DFP, physostigmine, X-substance (courtesy Col. Wood), and prostigmine. Larger injections produced uncoordinated hyperpneic activity with total ventilation diminished rather than increased. Torsal movements were greatly diminished while facial accessory respiratory contractions were simultaneously augmented. Whereas diminished torsal activity suggests lack of respiratory stimulation, intensified facial activity suggests powerful respiratory stimulation. It is consequently concluded all experiments gave evidence for central potentiation of nervous activity. Atropin given during the coordinated hyperpnea produced by anticholinesterase poisoning reduced all respiratory movements. In the more severe poisoning where torsal movements were diminished and facial movements augmented atropin simultaneously restored torsal movements and diminished facial movements. Atropin simultaneously augmented inspiratory and reduced expiratory components of torsal breathing. Anticholinesterases produced comparable differential effects on respiratory components.

Our results suggest that the principle of bi-polar function of neurons combined with reciprocal inhibitory interlocking of half-centers offers reconcilation of confusing differences of opinion on the theory of central humoral integration. In accordance with this working hypothesis four strategical regions are exposed to differential anticholinesterase activity—the excitatory and inhibitory poles, respectively, of antagonistic neurons. Coordinated hypercapnic hyperpnea could accordingly be attributed to symmetrical effects of increased *physiological* anticholinesterase activity. The uncoordinated hyperpnea produced by *foreign* anticholinesterases could similarly be assigned to a differentially increased anticholinesterase activity at the four cardinal regions of reciprocally coupled neurons. Imbalance of potentiation is regarded as an essential factor in the interpretation of anticholinesterase activity.

A comparison of the renal clearances of allantoin and inulin in man. M. FRIEDMAN and S. O. MYERS (by invitation). *The Harold Brunn Institute for Cardiovascular Research, Mt. Zion Hospital, San Francisco.* A preliminary study (Proc. Soc. Exp. Biol. and Med., to be published) indicated that when six human subjects were given allantoin by mouth, the latter's renal clearance was 123 cc. per minute. This suggested that the clearance of allantoin might be equivalent to that of inulin and as such, a measure also of the rate of glomerular filtration. Simultaneous allantoin and inulin clearances were performed on five normal males. Allantoin was administered by mouth and inulin by continuous intravenous infusion. The average allantoin clearance of these five subjects was 114 cc. per minute (Range: 96 to 123 cc.). The average inulin clearance was 108 cc. per minute (Range: 98 to 118 cc.). The allantoin clearance/inulin clearance ratio was 1.05 (Range: 0.98 to 1.17). In view of these results, it would appear that the renal clearance of allantoin may be used as a measure of the rate of glomerular filtration in man.

Treatment of non-specific ulcerative colitis for one year with intestinal extracts. M. H. F. FRIEDMAN, B. F. HASKELL (by invitation), and J. M. WALDRON (by invitation). *Department of Physiology, Jefferson Medical College, and Department of Surgery, Jefferson Hospital, Philadelphia.* Previously (Federation Proceedings, 6, 107, 1947) we reported that oral administration of extracts of hog's small intestine mucosa appeared to exert a beneficial effect in patients with non-specific ulcerative colitis. Treatment was based on the hypothesis that the disease resulted from a deficiency of an intrinsic protective factor normally present in the intestinal mucosa. Of 52 patients now under treatment 23 have been followed for a period of over one year. Only 2 of these 23 patients have failed to respond favorably to intestinal extracts. Improvement, commencing in 3 to 5 weeks, consisted of reduction in frequency of bowel movements, increase in stool consistency and disappearance of gross blood and mucus. Sigmoidoscopic evidence of mucosal healing was present usually after symptomatic improvement was noted. The lower rectal mucosa usually was last to heal. In cases with extensive involvement of the colon of long standing mucosal healing and symptomatic improvement were noted but radiologic examination did not show reversal of fibrotic changes in the colon structure. Withdrawal of intestinal extracts or substitution of a placebo were followed by relapse with remission occurring on resumption of treatment with intestinal extracts.

Isolation of the substance in the intestinal extract which is responsible for the beneficial effects in ulcerative colitis is greatly hindered by lack of an assay procedure using animals with similar lesions of the colon induced experimentally.

Preparation and assay of secretin. M. H. F. FRIEDMAN, and J. E. THOMAS. *Department of Physiology, Jefferson Medical College, Philadelphia.* In the method to be described, separation

of secretin from inert substances is achieved early in the fractionation procedures. Neither vacuum distillation nor adjustment of pH to maximum solubility or precipitation points is involved. Reproducible results by different workers in two separate laboratories have been obtained. The secretin prepared by this method is early freed from contamination by vasodepressor substances, pyrogens, cholecystokinen, enterocrinin, and pancreozymin. Preparations with a potency of 64 to 160 Agren cat units (16 cat units equals 1 dog threshold or 1 clinical unit) per milligram, assayed against a standard reference secretin by a procedure to be described, has been given intravenously to over 150 human subjects without any demonstrable reactions and has been found to be non-antigenic in both animals and man.

Renal function related to increased intra-abdominal pressure in anesthetized and unanesthetized dogs. DAVID M. FRENCH, PEDRO A. MOLANO and WALTER M. BOOKER (introduced by A. B. Luckhardt). *Dept. of Pharmacology, Howard University School of Medicine, Washington, D. C.* We have reported circulatory and respiratory effects brought about by acutely increasing the intra-abdominal pressure in anesthetized dogs, and effects on the urinary output and the non-protein nitrogen of the blood in chronic animals (unanesthetized) whose intra-abdominal pressure was increased gradually from week to week. No clearance studies were mentioned in these reports. In view of the marked increase in venous pressure in the abdomen and below in acute experiments and the evidence of kidney involvement in the chronic experiments (decreased urinary output, albuminuria, passive congestion demonstrated at necropsy), it was thought worthwhile to attempt to correlate the clearance efficiency of the kidney with various levels of intra-abdominal pressure, both in acute anesthetized (pentothal) and chronic unanesthetized animals. After urine and blood samples were collected, the latter from the femoral vein, 10 mgm. of creatinine was injected every 15 minutes in some instances, or every 10 minutes in others. Blood and urine samples were drawn at the same intervals. After a control period the intra-abdominal pressure was increased to 10, 20, 30, 40, and 50 mm. of mercury for half hour periods. We have observed that at low intra-abdominal pressure (10 mm. or less) there is no effect on the clearance of creatinine. At moderate intra-abdominal pressures (20 to 30 mm.), there is a gradual reduction in the clearance of creatinine and at higher levels, the clearance falls off to very low levels. In the chronic experiments the clearance of phenol-sulphonpthalein and of creatinine is dependent to some degree upon the speed with which the intra-abdominal pressure is raised from week to week. Evidence of adjustment to given pressures can be seen, provided increases in intra-abdominal pressure are not too rapid. At high intra-abdominal pressures (40 to 50 mm.), however, phenolsulphonpthalein and creatinine are significantly reduced in clearance rate.

The effect of rutin in experimental frostbite. FREDERICK A. FUHRMAN. *Department of Physiology, Stanford University School of Medicine, Stanford, California.* The extent of tissue loss and course of injury following freezing were studied in the feet of rabbits given the flavonol glycoside, rutin, and the results were compared with those obtained on control animals (J. Clin. Invest. 26: 229). Closely clipped hind feet of anesthetized rabbits were frozen for 3 minutes at −55°C. This severity of injury in 15 control animals invariably resulted in gangrene of the entire injured region; 11 animals subsequently lost all the frozen part, while 4 retained a narrow tongue of tissue on the plantar surface. Rutin was administered to 10 rabbits by stomach tube in doses of 50 or 100 mgm./kgm. daily. Six animals were pretreated with rutin for 1 to 10 days before frostbite, while 4 received rutin immediately following injury. The time required for development of wet gangrene was significantly longer in the rutin-treated animals than in the controls. In none of the rutin-treated animals was the extent of tissue loss as great as in control animals, and in most cases it was confined to the toes. Rutin delayed the loss of intravenously administered trypan blue into frostbitten areas of abdominal skin of rats. Recent results indicate that rutin in propylene glycol (50 mgm./kgm. i.v.) lowers the threshold concentration of epinephrine necessary to produce closure of capillaries in the rat mesoappendix. It seems probable that the effect of rutin in frostbite may be the result of an alteration in the pattern of local blood flow.

Effect of anoxia on contractility and metabolism of intestinal smooth muscle. R. F. FURCHGOTT (by invitation) and EPHRAIM SHORR. *Dept. of Medicine, Cornell University Medical College and The New York Hospital, New York City.* Contractions of segments of rabbit duodenum and jejunum were recorded with an isotonic lever. The medium at 37.5° was Krebs-bicarbonate 200 mg.% glucose. With 95% O_2-5% CO_2 bubbling through the medium, the tone, amplitude and frequency remained relatively constant for as long as 5 hours. On substituting 95% N_2-5%CO_2 after a control aerobic period, tone decreased markedly and contractions became sporadic and of irregular frequency and amplitude. On restoration of aerobic conditions recovery was manifested by return of tone to or above that of the control period, and restoration of frequency and amplitude. Failure of recovery was indicated by absence of increase in tone above the anaerobic level, and failure to resume the aerobic contraction pattern. On the basis of these criteria

intestinal segments exhibited good recovery after anaerobiosis lasting up to 120 minutes. Data will be presented indicating that glycolysis provides energy for the sporadic contractions and maintenance of viability during anaerobiosis. When intestinal segments were suspended in glucose-free medium and first allowed to deplete their substrate stores (Furchgott and Shorr, Proc. Soc. Exp. Med. Biol., **61:** 280 (1946)), they gave no contractions during a subsequent 60 minute anaerobic period, and usually failed to recover on the reintroduction of oxygen and glucose. The contractility studies with rabbit intestine agree well with metabolic experiments with strips of smooth muscle from dog small intestine. After 60–120 minutes of anoxia, dog smooth muscle showed good recovery of Qo_2 and high-energy phosphate stores on being returned to aerobic conditions.

Determination of the etiology of pathological effects of explosive decompression on the lungs of rats. MADELINE FUSCO, (by invitation), HENRY MELLETTE, (by invitation) and FRED A. HITCHCOCK. *The Laboratory of Aviation Physiology, The Ohio State University, Columbus.* In order to determine the etiology of the pathological effects of explosive decompression on lungs of rats, rats were: Group A—Explosively decompressed to 30 mm. Hg under an experimental schedule allowing 80% survival; Group B—Exposed to 97% nitrogen at ground level of such duration to allow 80% survival; Group C—Sacrificed without experimental procedure. The lung volumes of these rats were determined as described in previous publications. Grossly, the anoxia alone produced some edema and hemorrhage of the lung but this effect was much greater in rats explosively decompressed. Measurements of lung volume, lung weight, and lung air indicate that only part of the effects of explosive decompression on the lungs are due to anoxia. Studies are in progress to determine the actual degree of edema present and the composition of the edema fluid.

Peripheral vasodilator effect of a substance present in normal human urine. CLIFFORD G. GADDY (by invitation), HAROLD D. GREEN and J. MAXWELL LITTLE. *Department of Physiology and Pharmacology, Bowman Gray School of Medicine of Wake Forest College, Winston-Salem, North Carolina.* The intravenous injection of urine into dogs causes an increase in cardiac output, a decrease in peripheral resistance and a decline in mean arterial pressure. (Green, Little and Hester, Fed. Proc. **6:** 114, 1947). In the present study injections of urine were made into the femoral artery of heparinized dogs anesthetized with sodium pentobarbital. Venous outflow was recorded by a modified Gaddum technique (J. Physiol. LXVII. Proc. Physiol. XVI). Collateral venous return was prevented by a tourniquet. A constant mean arterial pressure was maintained with a "Lampson" pressure regulating bottle. Urine, collected and handled sterilely, was dialyzed in "Viskos" bags at 9.0°C. and stored at −20°C. Representative samples of urine were found pyrogen-free using the rabbit as test animal. All injections were 0.2 cc. of the urine preparation or dialyzed saline in 1.0 cc. of blood given in 20 seconds. The response was always noted with 20 seconds after injection started and lasted 60–100 seconds. Effects are expressed as per cent changes in total flow. Results: Plain unaltered urine: +41.5 to +131.6 (mean +62.7%); Seitz filtered urine: −24.1 to +132.8 (mean + 58.3%); urine dialyzed 24 hours in tap water and 24 hours in distilled water: +3.5 to +100.4 (mean +56.0%); urine, dialyzed after Seitz filtration: +30.7 to +104.5 (mean 34.4%); dialyzed saline: +10.6 to +34.8 (mean +32.6%). The peripherally acting vasodilator substance found in normal human urine is partly removed from urine by Seitz filtration, is not dialyzable and is not the result of bacterial contamination.

Exchange of carbon dioxide and oxygen in the nasopharyngeal part of the respiratory dead space. MORTON GALDSTON and SEYMOUR A. HORWITZ (introduced by J. Murray Steele). *Department of Medicine, New York University College of Medicine and Research Service, Third (New York University) Medical Division, Goldwater Memorial Hospital, New York 17, N. Y.* The exchange of carbon dioxide and oxygen in the nasopharynx was studied in two healthy adults at rest in a recumbent posture. Air samples were drawn at the end of inspiration through a small rubber catheter into an evacuated mercury gas sample tube. Precautions were taken to prevent contamination of the samples with room air and previously expired air. In 19 samples of pharyngeal air collected within 2 seconds after the end of inspiration there was an average rise of 2.8 mm. Hg CO_2 and fall of 2.6 mm. Hg O_2 as compared with inspired room air which contained 20.93 volumes per cent oxygen and 0.03 volumes per cent carbon dioxide. These values correspond to approximately 7 per cent of the carbon dioxide and 5 per cent of the oxygen tension change commonly believed to occur only in the alveoli. During breath-holding there was a progressive increase in pCO_2 and a generally slower decrease in pO_2. A prompt fall in the rate of gaseous exchange occurred after voluntary hyperventilation. There is reason to believe that the gas exchange occurs across the mixed glandular secretions which bathe the pharynx.

The results of these studies have a direct bearing upon such related physiological problems as the size of the pulmonary dead space based on carbon dioxide as compared with oxygen exchange, ratio of carbon dioxide production to oxygen consumption

(RQ) based on analyses of expired air and alveolar air, and the indirect method of calculating the level of mean effective alveolar air pO_2.

Studies of the pathological physiology of negative "G" in animals and man. J. L. GAMBLE (by invitation), R. S. SHAW (by invitation), O. GAUER (by invitation) and J. P. HENRY. *Aero Medical Laby., Air Materiel Command, Wright Field, Dayton, Ohio.* Increased pressure in the veins of the head from negative acceleration causes congestion and eventually petechial hemorrhages in the conjunctivae and in the mucous membranes of the accessory sinuses and middle ear. The vessels of the brain, surrounded by incompressible media in the "closed box" of the skull, do not rupture from short exposures. However, two subdural hematomas were observed in ten animals given four two-minute exposures to accelerations of negative 7 g repeated at short intervals. The increased venous pressure also causes edema in the cephalad portions of the body with a retrobulbar edema which may cause diplopia in humans. If the exposure is prolonged for more than five seconds, evidences of heart and central nervous system disturbance are seen. Electrocardiograms usually show bradycardia in both animals and humans. All degrees of heart block and various types of ectopic rhythms occurred in the dogs and interstitial myocardial hemorrhages were occasionally demonstrated. Blood pressure recordings from animals and humans manifesting these "vagal effects" show a fall in arterial and a rise in venous pressure causing a reduction in the arterio-venous differential pressure. Rabbit electro-encephalograms following exposures to negative g showed large slow waves and a depression of the normal activity. Four dogs died with respiratory failure out of the ten given four two-minute exposures to negative 7 g.

Effect of satiation on the intensity of the conditional and unconditional salivary secretion. W. HORSLEY GANTT. *Pavlovian Laboratory of the Phipps Psychiatric Clinic, Johns Hopkins Hospital.* The salivary conditional reflex (cr) depends upon many factors—kind of food, nature and intensity of conditional stimulus (cs) (Pavlov) and exponentially upon the amount of food—unconditional stimulus (Gantt). These definite relationships have been obtained under strictly controlled conditions—in a hungry animal. The following experiments were designed to show the effect of the state of hunger on the crs.

Dogs having stable secretory crs measured through a parotid fistula were used. After a daily series of readings of the salivary cr the dog was fed to satiation of food to which he was conditioned—500 to 2000 gms. After satiation the animal was negativistic toward food, and the cr fell immediately to zero. After some minutes it would eat a little food but the cr remained nearly zero. The UR showed no constant deviation. In four dogs (Kompa, Billy, Pat, Sechs) the cr before satiation averaged 0.666 cc.; after satiation 0.09 cc. (30 second readings); the UR 0.80 and 0.71 cc. respectively. The laws for cr intensity are seen only under rigidly controlled conditions; the most powerful factor is not intensity and kind of stimulus (cs or US), but the underlying emotional state of the animal—as the emotional tension decreases so does the conditional response, which may change within a few minutes from maximum to zero. The UR, however, maintains its level in spite of the changing emotional state. There is thus a sharp cleavage between cr and its corresponding UR.

Comparison of the effect of some steroids on the hearts of rats with choline deficiency. JOSEPH H. GAST and HAROLD L. DOBSON (introduced by Allen D. Keller). *Dept. of Biochemistry, Baylor Univ. College of Medicine, Houston, Texas.* Rat hearts were studied before and after 28 days administration of five representative steroids by planimeter measurements of the frontal cardiac area (FCA) from duplicate roentgenograms, leads II and III of the electrocardiogram and measurements of the blood pressure in the tail. Terminally the actual heart weight was compared to that calculated from final body weight by the equation of Rytand as used in previous studies (Fed. Proc. 4: 22, 1945). All animals with normal FCA initially showed no changes, a few with larger initial FCA showed decreases after injection of female sex hormones. Electrocardiographic studies showed heart rates to vary widely, but there was no axis deviation, and no change in PR interval, QRS, or in T waves, though P waves were found to be extremely labile. Contrary to some investigators no elevation in blood pressure was observed, however, any neurogenic effect may have been abolished by the pentobarbital used in all manipulations. Heart weights were uniformly low in all series. Microscopic examination of sections of adrenals, gonads aorta, kidney, liver and hearts demonstrated only the expected changes in the liver and gonads, except for non-specific fatty infiltration in the adrenal medulla of 7 of the animals.

Explosive decompression of monkeys at extreme altitudes. SAMUEL GELFAN and GEORGE D. DAVIS (by invitation). *Laby. of Physiology, Yale Univ. School of Medicine, New Haven, Conn.* The work on explosive decompression of rats as reported at the last meeting has been extended to monkeys. In addition to EKG and respiration, continuous recording was also maintained of blood pressure and EEG. The systolic and diastolic blood pressures were registered through an indwelling arterial needle connected with a Statham strain gauge pressure transmitter and recorded simultaneously with the other variables on a Grass electroencephalograph. As in the case of rats,

explosive decompression per se even up to 75,000 feet altitude does not kill the monkeys. When recompressed at free fall rate, the animals have survived explosive decompression to such altitudes in oxygen and at least 60,000 feet in air.

Studies of arterial oxygen saturation in patients with suspected arterial hypoxemia, with use of a modified oximeter. J. E. GERACI (by invitation), GEORGE E. MONTGOMERY, JR. (by invitation) and EARL H. WOOD. *Section on Physiology of the Mayo Clinic and Foundation, Rochester, Minnesota.* Resting arterial oxygen saturation in 27 patients with suspected arterial hypoxemia has been measured by Van Slyke analysis of samples of arterial blood and by photo-electric measurements (see abstract, Wood and Geraci) made on the ear of the patient at the time of withdrawal of the sample. The average resting arterial oxygen saturation as determined by Van Slyke analysis was 79.1 (13.4 to 98.2) per cent. The average difference between the photo-electrically determined arterial oxygen saturation and the results of Van Slyke analysis was 0.9 (−5.2 to 5.5) percentage points. Ninety-three per cent of the differences were within ±5 percentage saturation points. Multiple blood samples were obtained (see abstract, Wood, Montgomery, Geraci) from ten patients with cyanotic congenital heart disease. The average changes in arterial saturation in percentage points as measured photoelectrically and by Van Slyke analysis were: −4.0 and −3.3 after five minutes in the erect posture; −19.2 and −18.4 after walking two to five minutes at 1.7 miles per hour; and +9.2 and +9.5, respectively, after breathing 100 per cent oxygen for ten minutes. In a comparable series of eight patients, with the use of the Millikan CMR model 13 compensated circuit oximeter, the average decrease in saturation with exercise was 13.4 percentage points, as compared to the average decrease of 19.6 percentage points determined by Van Slyke analyses. Decreases in arterial saturation measured by the Millikan oximeter averaged 6.2 (+2 to −21) percentage points less than the results of Van Slyke analyses. In the modified oximeter group, this average difference was 1.4 (−2.9 to 4.9) percentage points. Figures in parentheses are extreme values.

Staphylocoagulase and staphylokinase. E. B. GERHEIM (by invitation), J. H. FERGUSON, and B. L. TRAVIS (by invitation). *Dept. of Physiology, Univ of North Carolina, Chapel Hill.* It has long been known that Staphylococci, particularly pathogenic strains, can coagulate oxalated or citrated plasmas and certain fibrinogen preparations. Smith and Hale (1944) showed the necessity for a cofactor in the coagulase reaction. The present study confirms the need for such cofactor and identifies it in a number of materials, especially the albumin fraction of plasma or serum. The staphylococcal agent, which we propose to call *pro*staphylocoagulase, is precipitated by established methods from chilled broth culture Staph. aureus filtrate, or centrifugate, by 3 vols. of cold 95 per cent alcohol. Its precursor nature is shown by failure to cause coagulation when added to bovine Plasma Fraction I (Armour's), whereas the fibrinogen is clotted to typical fibrin (dark-field microscopy) when cofactor is also supplied in the form of human serum albumin. In this system, lacking the precursor of the fibrinolytic protease (tryptase of plasmin), there is no clot lysis in 10 days at 37°C. However, human Plasma Fraction I (Harvard), previously shown to contain tryptogen (protease precursor), and currently found to have the coagulase-cofactor as well, is first clotted with prostaphylocoagulase and subsequently shows fibrinolysis. Thus, the staphylococcal product acts in a similar manner to streptokinase, in the lysis phenomenon only, although typically weaker. The term staphylokinase is appropriate for the factor responsible for this action.

The immediate pressor effect of desoxycorticosterone acetate in hypertensive and normotensive subjects. MELVIN L. GOLDMAN (by invitation) and HENRY A. SCHROEDER. *Department of Internal Medicine and the Oscar Johnson Institute, Washington University School of Medicine, and Barnes Hospital, St. Louis, Missouri.* Attempts were made to investigate the immediate effects in man or the intravenous injection of certain steroid compounds on the arterial pressure. Blood pressure was measured by a Hamilton optical manometer; blood content of the ear was estimated by a photoelectric plethysmograph; and cardiac output by the ballistocardiogram. The intravenous injection of 5 mg. of desoxycorticosterone acetate (dissolved in 2.5 cc. of propylene glycol) was followed by significant elevations of the blood pressure in hypertensive individuals. The responses observed lasted up to thirty minutes after injection. Similar effects did not occur in most normal individuals. Cardiac output, venous pressure, blood content of the ear, or electrocardiograms were not altered. The effects of other steroid compounds are being studied; among them progesterone was found to have a similar, although less powerful, action. From these results it would appear that desoxycorticosterone acetate can act as a pressor substance in some hypertensive individuals.

Calculation of effective afferent and efferent renal resistance. DOMINGO M. GOMEZ (introduced by H. W. Smith). *New York University College of Medicine.* Differential equations have been presented (La Revue Scientifique, No. 3272, May 1, 1947) defining hydrostatic pressure, blood flow and oncotic pressure in the kidney. The application of these equations to renal data involves knowledge of the locus at which filtration equilibrium is

reached. When capillary resistance is great, equilibrium is reached in the glomeruli; when it is small equilibrium may not be reached until the peritubular capillaries. The actual locus of equilibrium is unknown. Assuming that filtration equilibrium is reached in the efferent arteriole the following approximate relations may be drawn:

$$(1)\quad R^{(a)} = \frac{P_m - h_o^1 - H_0}{Q_o} = \frac{(R_a + b)\left(\frac{1}{E_G} + \frac{1}{E_R}\right) + R_a R_E}{\frac{1}{E_G} + \frac{1}{E_R} + R_E}$$

$$(2)\quad R^{(E)} = \frac{h_o + H_o - P_V}{Q_o} = \frac{(R_E - b + R_Z)\left(\frac{1}{E_G} + \frac{1}{E_R}\right) + R_Z R_E}{\frac{1}{E_G} + \frac{1}{E_R} + R_E}$$

$$(3)\quad F_f = \frac{q_o^1}{q_o} = \frac{R_E}{\left(\frac{1}{E_G} + \frac{1}{E_R} + R_E\right)(1 - C_o)}$$

where $R^{(a)}$ is the total resistance between the aorta and the locus of filtration equilibrium, and $R^{(E)}$ the resistance beyond this locus; P_m, mean arterial pressure; H_o, capsular pressure; Q_o, whole blood flow; h'_o, oncotic pressure at equilibrium; C_o, hematocrit; R_a, R_E and R_z the resistances of the afferent arteriole, efferent arteriole and collecting venule, respectively; E_G and E_R, permeability coefficients in the glomeruli and in the peritubular capillaries P_v, veinous pressure; b, locus of equilibrium relative to the efferent arteriolar resistance; q'_o, filtration rate; q_o afferent plasma flow; F_f, filtration fraction. Since $R^{(a)}$ and $R^{(E)}$ are series-parallel resistances proximal or distal, respectively, to the point of equilibrium, we designate them as effective afferent and efferent resistance.

The in vitro metabolism of testosterone to Δ^4-androstenedione-3,17 and cis-testosterone by rabbit liver homogenate. JOSE GONGORA (by invitation) and CHARLES D. KOCHAKIAN. *Department of Physiology and Vital Economics, University of Rochester, Rochester, New York.* In previous reports from this laboratory it has been demonstrated that testosterone is converted by rabbit liver slices to Δ^4-androstenedione-3,17 and cis-testosterone. It has been demonstrated now in three separate experiments that these changes also occur when the testosterone is incubated with a homogenate of rabbit liver prepared in a Waring blender. The products isolated accounted for 92 per cent of the original testosterone. The recovered material consisted of 87 per cent testosterone, 11 per cent Δ^4-androstenedione-3,17 and 2 per cent cis-testosterone.

The relation of the endocrine gland system to macrophagic activity. ALBERT S. GORDON and GRACE F. KATSH (by invitation). *Department of Biology, Washington Square College of Arts and Sciences, New York University.* In a previous communication, one of us (A. S. G., Fed. Proc. 5: 1946) reported a diminution in phagocytic activity of splenic macrophagic tissue following adrenalectomy in rats. In addition, administration of adrenal cortical extract, but not DCA, resulted in greater uptake of the colloidal agent (thorotrast). In the present work, 19 normal, 37 hypophysectomized and 7 starved rats were injected intravenously with 0.3 cc. thorotrast and killed 20 hours later. The spleens were then analyzed for thorium uptake. Seven of the hypophysectomized rats received 7 cc. of an adrenal cortical concentrate over a 2 day period prior to thorotrast administration; one cc. of cortical extract was given 9 hours after the injection of thorotrast. Six of the hypophysectomized rats received similar quantities of 1% saline solution. The remaining 24 hypophysectomized rats were untreated. The 7 started rats were restricted to a 5 grams of normal laboratory ration/day over a 32 day period which resulted in a 30% body weight loss. The mean values ± standard errors for thorium uptake/gm. dried spleen were the following: 1) normal untreated—43.3 ± 3.7 mg. 2) hypophysectomized untreated—51.2 ± 6.5 mg. 3) hypophysectomized + saline—38.1 ± 9.3 mg. 4) hypophysectomized + cortical extract—146.2 ± 14.9 mg. 5) normal starved—107.9 ± 12.7 mg. The thorium was observed to be localized largely in the macrophagic tissue.

Thus although hypophysectomy does not significantly affect phagocytosis by splenic macrophagic tissue, administration of cortical extract will greatly augment this activity. The question as to whether starvation, in increasing phagocytosis, acts through the adrenal cortex is being studied.

Acute hypothermia in guinea pigs. R. E. GOSSELIN (introduced by E. F. Adolph). *Dept. of Physiology, The University of Rochester, Rochester, N. Y.* Severe reductions of body temperature were produced in 30 unanesthetized mature guinea pigs by partial immersion in ice water. Measurements included colonic temperature, electrocardiograms, ventilation rate (minute respiratory volume), oxygen consumption, and CO_2 production. Regardless of the rate of cooling or rewarming, heart rates varied linearly with colonic temperature between 23° and 35°C., falling 18 beats per minute for each decrement of one Centigrade degree.

Below 23°C. the drop was erratic and often precipitous. Premature beats of auricular and ventricular origins, auriculo-ventricular and intraventricular blocks, and reversible ventricular fibrillation were all noted.

The early phases of cooling were characterized by increased struggling, hyperpnea, enhanced metabolism and cardiac slowing. When cooling at 0.2 to 0.3 degree per minute, maximal ventilation occurred ar 35°C. (33 to 37°) and averaged 180% of pre-immersion values. Maximal rates of oxygen consumption then averaged 170% of pre-immersion rates, or 250% of estimated basal values. Below 33°C., ventilation and oxygen consumption diminished progressively with temperature. Relative to the oxygen uptake, however, the ventilation rate remained high to the stage of complete apnea. Therefore the progressive fall in oxygen consumption cannot be ascribed to failure of external respiration.

Some animals recovered after the rectal temperatures were as low as 18°, others succumbing at temperatures as high as 22°. This large variation in lethal temperature may be due to uneven distribution of temperature. The mechanisms of hypothermic death remain obscure, but these data suggest that severe circulatory inadequacy often precedes respiratory failure.

Oxygen in bone marrow blood during prolonged hemorrhagic anemia. WILSON C. GRANT (introduced by Walter S. Root). *Department of Physiology, College of Physicians and Surgeons, Columbia University.* The purpose of this study was to observe *directly* the O_2 saturation of blood in red bone marrow during a period of constant and severe erythropoietic stimulation, after all reservoirs of preformed erythrocytes had been depleted. Adult dogs on stock diet were rendered anemic by repeated small hemorrhages. In 10 to 15 days this procedure lowered the Hb to 6 to 8.2 gm. per cent (O_2 capacity) where it was then maintained for 40 to 60 days by the removal of blood every 2 to 3 days. O_2 saturation measurements were made on arterial and jugular venous blood and on marrow blood obtained from the humerus. Control observations were collected during one to two weeks prior to bleeding. The determinations were resumed at weekly intervals 10–15 days after the start of the hemorrhages when the Hb level had reached the desired value. The dogs produced from 15 to 30 gm. Hb per week and remained in good condition despite the severe anemia. The reticulocyte percentage was increased while the saturation index decreased. Percentage O_2 saturation of marrow blood, although variable, showed no significant difference between the control and anemic periods. Similar results were obtained with arterial samples whereas jugular venous blood was less saturated, resulting in an increased A-V O_2 difference. From these findings it appears that vigorous erythropoiesis is not necessarily associated with a lowered O_2 saturation of the blood circulating through bone marrow.

The relation of initial blood pressure to adrenalin action. D. M. GREEN, A. D. JOHNSON, A. LOBB and G. CUSICK (introduced by R. Frederick Becker). *School of Medicine, University of Washington.* Fifty-one patients whose systolic blood pressures ranged from 90 to 246 mm. Hg were given a total of 80 infusions of adrenalin for period of 75 minutes' duration. Adrenalin was administered initially at an average rate of 0.2 micrograms per kilogram per minute. When the full effect had been obtained the rate was increased in steps of 0.2 mcg./kg./min. until the individual limit of tolerance was reached. The infusion was then maintained at this rate until the end of the period. Blood pressure and pulse rate were measured every 3 to 5 minutes during infusion and at increasing intervals for 12 hours afterward. Response to adrenalin was calculated in terms of net and unit changes in blood pressure, pulse rate and systolic-diastolic ratio. Individual variation was marked. However, when the patients were grouped according to initial pressure, no statistically significant mean differences in response were demonstrated. Post-infusion depression of blood pressure below the initial level was found to correlate more closely with the height of the initial pressure than with maximum pressure, maximum rise in pressure or maximum dose.

Vasodilation produced by etamon, priscol, body warming and spinal anesthesia in normal extremities. HAROLD D. GREEN and BEN C. OGLE (by invitation). *From the Department of Physiology and Pharmacology, Bowman Gray School of Medicine of Wake Forest College, Winston-Salem, North Carolina.* Temperature of the exposed skin of the toes, feet, shin, knees, thighs, fingers, forehead and room temperature were recorded with an 8 point Leeds and Northrup micromax over periods of 2 to 7 hours on patients in a constant temperature room (±1–2°C.) at environmental temperatures of 19–28° (av. 24°C.). Temperatures usually stabilized in 0.5 to 1.5 hours, at which time tetraethyl ammonium chloride (Etamon), 2 Benzyl 4,5 Imidazoline (Priscol), body warming or spinal anesthesia was begun. Body warming was by blanket and two 150 watt "reflector spot" lamps which strongly warmed the torso, but not the portions of the skin in which temperature was being measured. The most marked vasodilation was after spinal anesthesia. Etamon usually gave good vasodilation in doses of 500 to 1000 mg. given I.V. over 20 to 60 minutes. 10 mg. of Priscol was relatively ineffective, but 20 to 40 mg. often produced striking vasodilation. Body warming produced further vasodilation after etamon.

Studies of galactose and glucose metabolism. LAWRENCE GREENMAN and JOHN C. RATHBUN (introduced by T. S. Danowski). *From the Department of Research Medicine, University of Pittsburgh School of Medicine, the Renziehausen Foundation, and the Children's Hospital of Pittsburgh.* Galactose and glucose metabolism has been studied in a four month old white male infant with inability to handle galactose normally. The patient had cataracts, anemia, malnutrition, hepatomegaly, and ascites. In each of the studies a 50 per cent solution of galactose in water, 1.2 grams per kilo body weight, was injected intravenously during a 3 minute period. 15, 45, and 75 minute samples of venous blood were analyzed for galactose and total sugar. Glucose was then calculated from the two values. Prior to galactose restriction the 75 minute level of galactose in venous blood was found to be distinctly elevated (176 and 156 mgm. per cent). Glucose alone, insulin alone, and glucose and insulin together were then administered before the galactose and in each instance increased the rate of removal of galactose. The 75 minute levels were decreased significantly towards normal, but were still definitely above the values observed in control subjects. Alcohol did not affect the galactose tolerance. Standard glucose tolerance tests were unremarkable. However, in the galactose studies with insulin and with alcohol described above, glucose decreased to hypoglycemic levels. Galactose restriction over a four month period did not alter the galactose tolerance, but the child gained in height and weight, the liver decreased in size, the anemia and ascites disappeared, and the cataracts regressed markedly.

Effects of brain anemia on cortical oxygen consumption in vivo. R. G. GRENELL, P. W. DAVIES (by invitation), and D. W. BRONK. *Johnson Foundation, University of Pennsylvania.* Varying periods of anemia from fifteen seconds up to relatively long intervals, were induced in cats under dial anesthesia, by temporary compression of one common carotid artery following ligation of the second common carotid and both vertebral arteries. Readings of the rate of oxygen consumption before and after the anemia were made with the oxygen electrode which was placed over arterioles on the suprasylvian gyrus. Fifteen seconds of anemia induce no significant change in the oxygen consumption. Thirty seconds of occlusion result in an immediate but reversible depression of the oxygen consumption down to a factor of one half. The depression is followed by a period of steady recovery; both phases extending over a period of approximately twenty to twenty-five minutes. An anemia of one minute duration is followed not only by a much sharper drop in oxygen consumption, but also by a recovery period which lasts over a long period of time. More severe anemias produce irreversible depression.

Histological evidence has shown that permanent damage occurs in the cerebral cortex in as little as two minutes of complete anemia. The present findings indicate that severe depression of the oxygen consumption may result from one minute of anemia. Thus, in addition to observations of electrical activity changes and histopathology consequent to anemia of the brain, direct measurements of the oxygen consumption in vivo offer further evidence of the marked sensitivity of these cells to anoxia or arrest of the circulation.

Electrical correlates of psychiatric disturbances. R. G. GRENELL, B. MOORE (by invitation), H. S. BURR (by invitation), W. BROWN (by invitation) and S. FRIEDMAN (by invitation). *Divisions of Neuroanatomy and Psychiatry, Yale University School of Medicine and Fairfield State Hospital, Newtown, Conn.* In recent years it has been demonstrated that many physiological and pathological events produce measurable changes in the electrical properties of the organism. In many such instances, particularly in cases of psychiatric aberrations of the central nervous system, AC measuring techniques have yielded disappointingly little information of value. As a consequence of this, and in view of the interesting shifts in DC potential seen accompanying nerve injury and regeneration, it was deemed advisable to study DC potential differences in psychiatric patients under various conditions. With the Burr-Lane-Nims DC microvoltmeter and reversible non-polarizable silver-silver chloride electrodes, a standard series of 8 measurements between areas on the head and chest was made in 61 controls (non-hospitalized, random individuals) and 150 psychiatric cases, over 100 of which were diagnosed as schizophrenics. In a relatively small group of patients, readings were made throughout a course of electric shock treatments; in others, readings were made before, during and after insulin shock. Two criteria were used in analyzing the data: a) arithmetic sum—the sum of the 8 potential differences in millivolts without regard to sign, and b) range—the difference between highest positive and lowest negative reading in each set of measurements.

Although the investigation is in its most elementary stages, and many variables have therefore not been taken into account (age, sex, etc.) the data thus far obtained appear to be of enough significance to justify further detailed studies of both clinical and fundamental physiological nature. There appears to be a definite correlation between the measured potentials and the psychiatric status of the individual (control sum mean = 11; schizophrenic sum mean = 65 . . . control range is + to − 5; patient range as far out as + 30 to − 30)

In other words, the method demonstrates electrical correlates of psychiatric conditions which are of quantitative nature. In successfully treated cases, electric and insulin shock have shifted the abnormal potential levels of the patients back to those of normals. The fact that insulin pushes abnormally high potentials down into the normal range, gives suggestive evidence of an important physiological nature, namely, the relationship between potential levels and metabolism.

A simple, relatively rapid test appears to be at hand for use in the clinic as an aid to psychiatric diagnosis and evaluation of therapeutic procedures. It is also felt that the observations present an important clue to the possible physiological nature of certain psychiatric conditions, as well as suggesting an extremely provocative relationship between DC potential and metabolic levels.

Hepatic lymph and ascitic fluid following experimental chronic obstruction of the inferior vena cava. J. H. GRINDLAY, E. V. FLOCK and J. L. BOLLMAN. *Division of Experimental Medicine, The Mayo Foundation, Rochester, Minnesota.* In other studies of hepatic lymph we observed greatly increased flow in dogs with carbon tetrachloride cirrhosis of the liver accompanied by ascites. Our experiments suggested the possibility that ascitic fluid is the result of venous congestion in the liver and that this fluid comes largely from the liver or the extrahepatic liver lymphatics. Accordingly, in order to produce a slowly progressive venous congestion in the liver, we wrapped cellophane bands loosely about the inferior vena cava above the diaphragm in a series of dogs. Six to eight weeks later these dogs had ascites, greatly enlarged and dilated liver lymphatics, and large, granular livers. Although the liver in each case showed venous engorgement, the spleen, portal veins and mesenteric lymphatics were not engorged. The rate of flow of hepatic lymph was greatly increased in these dogs and each had two to three liters of free fluid in the peritoneal cavity. Both lymph and ascitic fluid were quite clear, the former faint yellow, the latter faint pink. Plasma protein and cholesterol concentration in most dogs were slightly low. Plasma phospholipid was normal and alkaline phosphatase, although there was obvious liver damage, was not elevated. Protein, cholesterol, phospholipid, and alkaline phosphatase concentrations were lower in hepatic lymph than in plasma and lower in ascitic fluid than in lymph.

Relationship of volumetric rate of blood flow to arterial diameter. RICHARD A. GROAT. *Department of Anatomy, The Bowman Gray School of Medicine of Wake Forest College, Winston-Salem, North Carolina.* It was desired to find if, (*1*), $Q = kD^n$. Q is flow-rate to which the prevailing inside diameter, D, of an artery is adapted; k and n are constants. The validity can be tested by measuring arteries participating in dichotomizations, calculating for each branching the *equating value of* n, (n_{eq}), in $D_a^n = D_b^n + D_c^n + \ldots +$ D_s^n where a signifies parent vessel, the other subscripts, daughter vessels. If n_{eq} be found constant, then n in $Q_a = kD_a^n$, $Q_b = kD_b^n$, etc., will be the same for all dichotomizations. Each daughter is in turn a parent; k will be the same for all dichotomizations. Some variation of n_{eq} will not negate suitability of (*1*), since biological processes involved in adaptation of D to Q might be expected to lack precision. Furthermore, variation in n_{eq} may be much greater than corresponding variation in n in (*1*). Vessels from 24 to 3200 microns were recorded; the pulmonary circulation and aortic arch branchings were excluded. The mean of n_{eq} was 2.6, which established the mean of n as 2.6. Extremes of n_{eq} were 2.1 and 3.1. Greatest discrepancy of D's at a given dichotomization from the calculated diameters necessary for n_{eq} to equal 2.6, was 3.18%. Calculated limits of n, with D in microns, were 2.6 ± 0.02. Thus, $Q = kD^{2.6}$. Invoking Poiseuille's law, we obtain $(P_1 - P_2)/L =$ k.u. $1/D^{1.4}$, which, even with due consideration to decrease in viscosity, u, in tiny vessels, shows that pressure drop per unit length is always greater in a daughter than in its parent.

Hepatic arterio-venous oxygen differences in patients with normal and diseased livers. A. E. GROFF, (by invitation), E. DEF. BALDWIN, (by invitation) and S. E. BRADLEY, *From the Department of Medicine, College of Physicians and Surgeons, Columbia University, and the Presbyterian Hospital in the City of New York.* Simultaneous collection of hepatic venous blood (by venous catheter) and arterial blood were made during the determination of estimated hepatic blood flow (EHBF) by Bromsulfalein clearance in nine normal human subjects and in five patients with portal hypertension due to cirrhosis. Normally, the oxygen A-V difference average 3.4 ml. per cent, ranging between 2.5 and 4.5 ml. per cent. The splanchnic oxygen consumption averaged 50.9 ml. per min. and varied between 38.6 and 68.2 ml. per min. When hepatic blood flow was reduced by disease (5 patients with portal hypertension), the average A-V difference was 5.3 ml. per cent (3.4 − 7 ml. per cent). This increment was probably not attributable to an increased proportion of portal venous blood in the hepatic venous outflow as a result of the elevated portal pressure, since in one patient with Banti's syndrome due to a splenic arteriovenous fistula who had a normal liver at necropsy, EHBF and the hepatic oxygen (A-V difference) were within normal limits. The portal venous contribution was eliminated in another patient by ligation of the portal vein at the time of porto-caval anastomosis two years prior to study. In this instance, an EHBF

of 460 ml. per min. was associated with an oxygen A-V difference of 6.3 ml. per cent, despite an exclusively arterial hepatic blood supply. Thus, it appears that the hepatic oxygen A-V difference in man is, in part at least, a function of the rate of hepatic blood flow.

Inhibition of gastric secretion by "histaminase". M. I. GROSSMAN, C. R. ROBERTSON (by invitation), and W. L. ANDERSON (by invitation). *Dept. of Clinical Science, Univ. of Illinois College of Medicine, Chicago.* Previous attempts to counteract histamine stimulated gastric secretion by injection of histaminase preparations have been unsuccessful. A new, more highly purified and less toxic histaminase preparation (Winthrop) was used in the present studies. This material, prepared from hog kidney, contained approximately 1 Winthrop unit of histaminase activity per milligram of dry powder. Intravenous injection of 50 units per kilogram into dogs with gastric pouches receiving 0.0125 mg. of histamine dihydrochloride subcutaneously every ten minutes resulted in 90 to 100 per cent inhibition of the gastric secretory response for from 1 to 6 hours. The gastric secretory response to food and to parasympathomimetic drugs was similarly inhibited by the histaminase preparation. Purification of the product resulted in a material free of emesis-producing and vasodepressor substances. Although the gastric secretory depressant is destroyed by boiling, it is not yet certain whether the depression of gastric secretion is due to the histaminase or to some other constituent of the extract.

Preganglionic B fibers to the inferior mesenteric ganglion of the cat. HARRY GRUNDFEST and DOMINGO AMPIL (by invitation). *Dept. of Neurology, College of Physicians and Surgeons, Columbia Univ., New York.* The hypogastric nerve of the cat contains both B and C fibers, whereas the preganglionic nerves to the inferior mesenteric ganglion have A and B components. The C fibers of the hypogastric nerve therefore are clearly postganglionic, but the nature of the B fibers in the nerve has been uncertain. Oscillographic techniques with graded antidromic stimulation of the hypogastric nerve in intact or excised ganglionic preparations shows that most, if not all, the B fibers of the nerve are preganglionic. Their electrically evoked response ascends to the ganglion, and stimulates the latter, as is evidenced by a response descending the nerve in C fibers. The origins of the respective electrical activities are evidenced by the phase of the ascending and descending potentials. Preganglionic B fibers thus can make synaptic connections with cells in more than one autonomic ganglion by terminations *en passant*.

Diet and gastro-intestinal symptoms at altitude. M. MASON GUEST. *Wayne University, College of Medicine.* The effect of a bland diet upon the incidence of gastro-intestinal symptoms during a 3 hour sojourn at a simulated altitude of 38,000 feet was tested in a group of 2,448 healthy men between the ages of 18 and 30. The diet, consisting of fruit juice, cereal (bran excluded), white toast, jam or jelly, lean beef or fish, baked or boiled potato and milk or tea was fed at meal time during the 24 hour period preceding the decompression. The subjects, under supervision at all times, were not permitted to eat between meals or drink carbonated beverages or milk shakes. The control group was obtained by random sampling of the military population which furnished the experimental subjects. These individuals were not warned of the possible effect of diet upon gastro-intestinal symptoms and were provided with army garrison rations with no restrictions on between meal feeding. The control sample consisted of 1,915 men.

Symptoms	Control	Experimental
	per cent	*per cent*
Total gas pains	6.6	6.7
Severe gas pains*	1.9	2.4
Total nausea and vomiting	4.0	3.7
Severe aero embolism*	11.7	13.3

* Recompression because of severity of symptoms.

Since the bland diet had no effect in promoting a significant reduction in the incidence of symptoms, a re-evaluation of the effect of diet upon gastro-intestinal irritability may be warranted.

The quantitative effect of hemorrhage on plasma proteins. C. C. GUTHRIE and MARIAN E. LEE (by invitation). *Dept. of Physiology and Pharmacology, School of Medicine, Univ. of Pittsburgh.* These results were obtained on blood samples removed from six etherized dogs during measurement of their blood volumes by a modified Welcker method. In three, the proteins were calculated from specific gravity of serum; but in the others they were also determined by heat coagulating, drying, extracting with chloroform and ether and with water. The results by the two methods did not significantly differ. About forty minutes after the first sample was drawn, a total quantity equivalent to about 40% of the original blood volumes was removed in 7 samples. At this time, the circulating and removed proteins showed a percentile increase over the original amount of from 7.9 to 16.6, averaging 11.48. Following this, bleeding was continued and 0.9% NaCl solution intravenously injected until death, which occurred about thirty minutes later, at which time the protein total increase was 28.4 to 39.4%, averaging 34.6. In three experiments, fibrin was determined by whipping the blood samples, washing, drying and weighing. After hemorrhage only, there was an increase of

14.6 to 43.7%, averaging 29.1. At death, the total increase was 41.6 to 70.5%, averaging 60.2.

Resistance characteristics of rectifier element in single nerve fibers. RITA GUTTMAN. *Brooklyn College, Brooklyn, N. Y.* In 1941 it was shown that the nerve fiber membrane acts as an electrical rectifier, permitting current to flow more easily outward than inward. The rectifying property was found to decrease reversibly on narcotization or treatment with K and to disappear on death, when the membrane acts as an ohmic resistance. The present study investigates the resistance characteristics of the rectification curve in an attempt to discover whether the anodal or cathodal portion or both are affected on loss of rectification.

One end of a single nerve fiber of the squid was dipped into isosmotic KCl, killing that end and removing its rectifying properties. The other end was placed in sea water or experimental solution. The interelectrode stretch was in oil. Resistance offered to an E.M.F. of varying magnitude sent into the fiber first in one direction and then in the other was measured by means of a direct current Wheatstone bridge and calculated currents were plotted against applied voltages. Anodal currents and voltages were termed positive and cathodal negative. Plotting voltage against current, the experimental curve deviates more from the straight line which would represent an ohmic resistance in the first quadrant, where current and voltage are positive, than in the third quadrant, where current and voltage are negative. On narcotization, treatment with K or death, the anodal portion of the curve changes more than the cathodal as a straight line is approached. As time elapses, excitability is lost first. Then the resting potential and rectification concurrently disappear.

Glutamic acid in neural activity. C. HABER, L. SAIDEL, (introduced by R. W. Gerard,). *Dept. of Physiology, The University of Chicago.* A microbiological method has been applied to the determination of free and combined non-protein glutamate in nervous tissue. The tissue is homogenized and proteins precipitated with a minimum of tungstic acid. Samples of the supernatant are assayed, with and without prior acid hydrolysis, by addition to a modified Snell and Wright medium (J. Biol. Chem., 1944, **151:** 193; 1941, **139:** 675) adequate, for growth of Lactobacillus 17-5, except in its glutamate content. These mixtures are inoculated with the bacteria and the 72 hr. turbidities, read with a photoelectric colorimeter, compared to those obtained with standards of glutamate-medium mixtures. Brains of rats killed by decapitation contain about 125 micrograms free glutamic acid and 220 micrograms total non-protein glutamic (free and combined) per 100 mgms. tissue, wet weight (standard error ± 4 micrograms). Preliminary results indicate that strychnine convulsions produce approximately 30% decrease in free or combined glutamic acid in brain. Effects of other changes of neural function and metabolism on glutamate concentration in brain and nerve will be discussed.

The phosphatide composition of human erythrocytes. M. H. HACK (introduced by Hubert R. Catchpole). *Dept. of Pathology, Univ. of Illinois, College of Medicine, Chicago.* It was recently shown (J. Biol. Chem. **169:** 137, 1947) that the phosphatide composition of human erythrocytes could be determined indirectly. The data of the present report was obtained by direct assay, and confirms the results previously obtained. Erythrocytes were obtained by centrifugation of citrated blood and the cells washed repeatedly with physiological saline and dried by (1) lyophilization (2) acetone. The dried cells were extracted with chloroform-methanol and the extracts assayed for the component phosphatides by the method previously described. The data is expressed in mM/liter of cells calculated from the dry weight equivalent, which was found to be approximately 360 grams per liter of wet-packed cells.

Phosphatide	Lyophilized		Acetone dried
	Direct	Indirect	
Total	3.82	3.91	1.07
Cephalin	1.57	1.72	0.51
Lecithin	1.15	1.13	0.19
Sphingomyelin	1.10	1.06	0.37

The agreement between the two methods, using lyophilized cells, is evident. Drying by acetone extraction decreases the phosphatide content considerably, particularly the lecithin fraction. It was found that lyophilized erythrocytes swell tremendously and form a tough, rubber-like mass when wetted with chloroform-methanol; this mass could, however, be broken up in a Waring Blendor to facilitate the preparation of the extract. Although acetone-dried cells did swell slightly, they did not agglutinate.

The brightness threshold as a function of area and receptor number in various retinal regions. C. HAIG and E. M. HAIG (by invitation). *Dept. of Physiology and Biochemistry, New York Medical College, New York 29, N. Y.* Measurements of the minimal brightness (I_t) required for stimulation of dark-adapted rods and cones were made, over a range of stimulus areas (A) of $1/10^7$, in retinal regions from the center to 18° in the temporal periphery. For rod measurements the eye was completely dark-adapted. For cones the eye was first light-adapted, and measurements were made during dark adaptation in the period when cones are completely dark-adapted but rods are still "nonfunctional". Wavelengths longer than 555 mμ were

used to avoid differential absorption of the violet end of the spectrum by the yellow macular pigment. To compute rod and cone numbers, Østerberg's (1935) counts of a single retina were employed. The following quantities for one eye are approximate. For small areas, $I_t A$ = Constant in all retinal regions. For rods the relation holds rigorously for areas of subtense $< 0.5° - 0.8°$ (2800 rods), and for cones for areas of subtense $< 0.2° - 0.5°$ (600 central and 80 peripheral cones). Within these limits, for a constant visual effect (given threshold value) in all peripheral regions, the number of receptors is constant. Thus, for a constant, small area, I_t is inversely proportional to the receptor population density, i.e., to the concentration of photosensitive material. For larger areas, I_t approaches a constant minimal value (I_{min}) such that $(I_t - I_{min}) A$ = Constant. The data indicate that the brightness limin is inversely proportional to the exposed receptor surface area, summation of subliminal nerve impulse frequencies probably occurring when more than one receptor is exposed.

Industrial efficiency as affected by various kinds of foods consumed during the mid-morning and mid-afternoon rest periods. JOHN HALDI and WINFREY WYNN (by invitation). *Dept. of Physiology, Emory University, Emory University, Georgia.* This investigation was conducted in a large cotton bag factory. All the subjects were women. They were engaged continuously throughout the working hours of the day sewing bags on electrically driven sewing machines. Their compensation was determined by the number of bags sewed. An arrangement of this wort has the following advantages: 1. it provides a monetary incentive for maximum production; 2. the hourly output can be accurately determined by counting the number of bags sewed. On different days of the week the refection was as follows: (1) cake, banana, milk; (2) frankfurter, bun, potato chips, soft beverage; (3) chocolate bar; (4) soft beverage only; (5) nothing. Only rough approximations were made of the caloric intake, but it is quite obvious that there was a marked difference in the amount and kinds of food eaten on different days. The experiment extended over 14 consecutive working days. The last scheduled experiment with no refection was cancelled because of the complaint registered by the subjects. Owing to the wide variation in the work output from one subject to another, observations were discarded unless the subject took part in all the experiments of the week.

From the data obtained in 52 individual observations after each type of refection, it is concluded that work output was not affected by the kind or amount of food eaten during the mid-morning and mid-afternoon rest periods. Abstention from food during the rest periods on the two days in which this experiment was conducted likewise had no effect on work output.

Studies on the mechanism of "acid rebound" in gastric acidity. E. HALE (by invitation) and M. I. GROSSMAN. *Department of Clinical Science, University of Illinois, College of Medicine, Chicago.* One possible mechanism of the alleged "acid rebound" in gastric acidity following the administration of sodium bicarbonate is metabolic alkalosis. The effect of 200 cc. of 2.6% sodium bicarbonate, when given intravenously, was observed in 12 experiments on 5 total pouch dogs. Secretion was established and maintained by repeated injections of 0.025 milligrams of histamine dihydrochloride every 10 minutes. There was no significant change in the average gastric secretion during the two hours of observation following a two hour control period and one hour of infusion. The infusion of 1.8% sodium chloride in 10 control experiments demonstrated a moderate depression, in most instances, as compared with the control level of secretion. Another effect of the oral ingestion of sodium bicarbonate is the liberation of carbon dioxide. The possibility that carbon dioxide gas could shorten the emptying time of the stomach or stimulate secretion by distention or by a local effect on the parietal cell was studied. Two experiments were performed on each of 10 normal, fasting subjects. Two hundred fifty cc. of carbonated or uncarbonated distilled water were given alternately during the first or second hour following a 30 minute basal period. Carbonated water had no consistent or significant effect upon emptying time at 15 or 25 minutes or on the secretion of free acid. It is concluded that if "acid rebound" does occur it is not due to the changes in systemic acid-base balance or gaseous distention.

The influence of substances affecting body temperature on thermal polypnea. V. E. HALL, R. GRANT (by invitation) and J. FIELD, *Department of Physiology, Stanford University, California.* In a study designed to determine whether the metabolic activity of the central nervous system is a factor determining the level at which body temperature is regulated, the effect of certain substances affecting body temperature on (a) oxidative and glycolytic activity of cerebral cortex (see abstract by Field, Peiss and Hall), and (b) the respiratory response to heat (thermal polypnea) were determined. Typhoid-paratyphoid vaccine, in doses evoking a fever of 1–2°C., raises the rectal temperature level at which thermal polypnea appears by about 2°C. If the rabbit is panting before injection, panting stops within fifteen minutes, but can be restored by raising the body temperature. The respiratory response to the inhalation of 95% O_2-5% CO_2 is not depressed by vaccine, so that the effect is presumably not on the inspiratory center. Persistence of normal rhythmic

respiration in vagotomized rabbits shows that the pneumotaxic mechanism is not inactivated by the vaccine. Magnesium chloride (3 mM per kg. intraperitoneally) evokes a polypnea in rabbits at an environmental temperature of 20°C. or higher, which does not appear at 10°. At both temperatures, the rectal temperature is reduced by 1–2°C. These facts, together with the ear vasodilatation evoked, suggest that magnesium chloride reduces the body temperature level at which heat defense mechanisms are activated. Similar studies of the effects of antipyrin, dinitrophenol and other substances affecting the body temperature are under way.

Changes in radial arterial blood pressure during surgical resection of coarctation of the aorta. G. A. HALLENBECK, E. H. WOOD and O. T. CLAGETT (by invitation). *Section on Physiology and Division of Surgery, Mayo Clinic, Rochester, Minnesota.* A mobile recording oscillograph which, with its associated pick-up units, can be transported to the operating room and used there without interfering with surgical procedures, has been designed to permit recording of physiologic data on patients undergoing operation. In its present form the apparatus can record arterial and venous blood pressure, using hypodermic strain gauge manometers; respiratory pressure changes within the anesthesia mask, using a similar strain gauge manometer with an air system; arterial oxygen saturation, using Millikan's compensated circuit oximeter; the electrocardiogram and the pulse rate, using a recording cardiotachometer; and appropriate time signals. The relatively large size of the instrument provides considerable flexibility and other recordings can be added as desired. Records were obtained of radial blood pressure changes during four surgical resections of aortic strictures. In two cases in which the left subclavian artery was temporarily clamped, severe acute hypertension occurred. In two cases in which the aorta was clamped proximal to the coarctation but distal to the left subclavian artery, no significant alteration in radial blood pressure resulted. With gradual opening of the vascular anastomosis which re-establishes aortic blood flow after resection of the stricture, radial blood pressure fell smoothly for a period of thirty to ninety seconds. The decrease in radial systolic blood pressure at this time ranged between 30 and 90 mm. of mercury.

Comparison of the Fick and Dye injection methods of measuring the cardiac output in man. W. F. HAMILTON, R. L. RILEY (by invitation), A. M. ATTYAH (by invitation), ANDRÉ COURNAND, D. M. FOWELL (by invitation), A. HIMMELSTEIN (by invitation), R. P. NOBLE (by invitation), J. W. REMINGTON, D. W. RICHARDS, JR. (by invitation), N. C. WHEELER (by invitation), and A. C. WITHAM (by invitation). *Department of Physiology, University of Georgia School of Medicine, Augusta, and the Chest Service, Bellevue Hospital, Columbia University, New York.* The cardiac output was measured in a series of 48 experiments using the direct Fick procedure by means of intravenous catheterization. This was followed immediately by a determination using the dye injection method. The work was carried out in two laboratories, 18 comparisons being made in the Georgia laboratory and 30 in the Columbia laboratory as a result of a cooperative project in which two of the Georgia group (J. W. R. and W. F. H.) worked with the Columbia group in their laboratory. There was a wide variety of experimental conditions including normal subjects at rest and during exercise, and patients with mild congestive failure and various pulmonary abnormalities. The oxygen consumption varied from 182 to 1660 cc. per minute, and the cardiac output varied from 2.5 to 16.8 liters per minute. The distribution of the results about the line of identity was almost symmetrical, so that the average of all the determinations by one method was nearly the same as by the other. The average scatter, 14%, was no greater than would be expected when the known inaccuracies of both methods are considered. The data include oxygen consumption, circulation time, mean circulation time, blood volume, volume of blood in heart, lungs and great vessels, as well as the cardiac output.

Possible influence of cosmic energy cycles on growth. FREDERICK S. HAMMETT. *The Lankenau Hospital Research Institute, Philadelphia.* Abbot (Science, **105**, 632, 1947) reports a solar constant of radiation with periodicity of 6.6456 days: invariable, but with terrestrial effects subject to phase displacement of 1 or 2 days. There is week-to-week rhythmicity in extent of expression of basic growth and life-cycle activities (initiation; proliferation; differentiation; organization; maintenance; regression; catabolism) in the marine hydroid Obelia (457198 hydranths; 35093 gonangia) during a 24-hour time-slice in culture. The similarity of the rhythmicity cycle to that of solar radiation suggests it may be the product thereof. There is no open evidence rhythmicity is greatly conditioned by endogenous factors; or exogenous which are not derivatives of the solar cycle. Some valid interactivity difference exist. Rhythmicity of differentiation is less than that of organization or proliferation. Differentiation is cell specialization; organization is cell segregation; proliferation is cell multiplication. Each is expression of chemical processes specific and peculiar thereto. The differentials suggest processes concerned in cell specialization are more resistant to cosmic energy cycles than are those of cell segregation or cell multiplication. This fits the probabilities. For cell specialization is primary to ontogenetic growth,

and the most basic activity should be least sensitive to interference from exogenous factors. These observations are evidence that proper evaluation of the effects of cosmic (or atomic) energy on growth depends on knowledge, not of the reaction of growth-as-a-whole, but rather on knowledge of the differential reactions of the components of growth.

Relationships between the cerebral and cerebellar cortices in the cat. JOHN L. HAMPSON (introduced by C. N. Woolsey). *Department of Physiology, The Johns Hopkins University School of Medicine, Baltimore 5, Md.* The projection of functional areas of the cerebral cortex onto the cerebellar cortex has been studied in the lightly anesthetized cat by means of the evoked potential technique. Using sodium pentobarbital as the anesthetic agent, the stimulating electrodes were applied to those points on the cerebral cortex from which the largest responses were obtained by stimulation of the appropriate peripheral receptor. The cerebellar cortex was simultaneously explored and the evoked cortical potentials, as observed by oscillographic methods, were recorded. The results show that: 1. Auditory area I projects to the entire tuber vermis. 2. Auditory area II (Woolsey and Walzl) projects to the entire tuber vermis. The latency of the evoked potential in this instance is somewhat less than for the auditory area I system. 3. Somatic area I (the post-central homologue) projects to the contralateral half of the anterior lobe. 4. Somatic area II (Adrian, Woolsey) (i.e. the ant. ectosylvian gyrus) projects to the tuber vermis, the pyramis and to the contralateral paramedian lobule. 5. The cortical autonomic center for the eyes on the medial surface of the frontal pole (Siebens and Woolsey) projects to the contralateral cerebellar hemisphere (both Crus I and II), to the contralateral lobus simplex and to the contralateral half of the culmen, with the maximal responses occurring in the medial folia of the contralateral cerebellar hemisphere.

The cerebral cortical areas studied show considerable overlap in their cerebellar representation.

Physiological adaptations to anoxia in congenital heart disease with cyanosis. J. C. HANDELSMAN (by invitation), RICHARD J. BING, L. D. VANDAM (by invitation). *From the Department of Surgery, The Johns Hopkins University and Hospital.* The arterial oxygen saturation of individuals with congenital heart disease and cyanosis is comparable to that of normal persons at altitudes ranging from 5000 to 30,000 feet. In both groups adaptations are directed toward restoring normal tissue pO_2 (Barcroft), by decreasing the gradient between the pO_2 of inspired air and the mean capillary pO_2. At high altitudes the atmospheric-arterial pO_2 gradient is reduced primarily by hyperventilation. The gradient from arterial pO_2 to mean capillary pO_2 (tissue pO_2) is reduced principally because of the shape of the hemoglobin dissociation curve and to a lesser degree by an increase in the oxygen carrying capacity of the blood and in the cardiac output (Houston and Riley). In tetralogy of Fallot the cause of anoxemia is the intracardiac shunt rather than reduced barometric pressure. Thus, reduction of the gradient between the pO_2 of inspired air and alveolar air is useless, and the only effective measures toward restoring normal tissue pO_2 consist in reducing the gradient from arterial to mean capillary pO_2. Again the shape of the hemoglobin dissociation curve is most important, the increased oxygen carrying capacity of the blood much less so. The anatomical lesion limits the effect of the systemic flow. In contrast to normal oxygen uptake observed in persons at high altitudes, a lower basal metabolic rate is seen in patients with tetralogy of Fallot. The serum CO_2 and pCO_2 are low; the blood pH normal. This compensated CO_2 deficit contrasts with the respiratory alkalosis seen at high altitudes.

The failure of intravenous glucose to inhibit food intake in dogs. M. E. HANSON (by invitation) and M. I. GROSSMAN. *Dept. of Clinical Science, Univ. of Illinois College of Medicine, Chicago.* Experiments were performed on four mature dogs, standardized as to weight and daily food intake. The diet consisted of constant proportions of a dry commercial dog food mixed with cooked meat juice, and it was given ad libitum for a 45 minute period at the same time each day. In one series of experiments from 200 to 400 cc. of a sterile 20% glucose-0.3% NaCl solution were injected intravenously in one hour, daily for seven days. In other experiments a 15% glucose-0.3% NaCl solution was administered over a four hour period for three or more days; volumes in these instances depended upon the capacity of dogs to metabolize glucose (1.7 gms./kg./min.) and ranged from 140 to 235 cc. per hour. As a control for each experiment, equal volumes of an 0.85% or 0.6% NaCl solution were injected for similar lengths of time on days either preceding or alternating with those for glucose injection. All injections were concluded twenty minutes before feeding the animals. We observed no increase in rectal temperature or signs of restlessness or nausea which would suggest reaction. Decrement of food intake following injections, when it occurred, was of the same magnitude whether saline or glucose had been administered. In no case did the decrement exceed 25% after glucose administration, which change was within the range of spontaneous fluctuation of control food intake. Furthermore this decrement decreased with successive days of injection, presumably due to adjustment of the animal to the experimental procedure.

Experimental investigation of pain intensity in labor. JAMES D. HARDY and CARL T. JAVERT (by invitation). *From the Russell Sage Institute of Pathology, The New York Hospital, and the Departments of Physiology, Gynecology and Obstetrics, Cornell University Medical College, New York, N. Y.* Using the method of estimating the intensity of the pain of labor by comparison with a pain of known intensity, measurements were made on ten women during the first and second stages of labor. Four of these patients received no analgesic. During the early part of the first stage of labor the pain intensity increased from three dols to five dols. In the later part of the first stage pain intensity attained a moderately severe level of approximately seven to eight dols. In the second stage of labor ceiling pain was experienced (ten and one-half dols) during the period of uterine contraction.

The effect of certain common analgesics upon the pain intensity in the first stage has been studied and it was observed that although no pain threshold raising effect was achieved by the use of moderate amounts of the analgesics a marked decrease in the intensity of labor pain was produced for short periods following administration of the agent. As the decrease in pain is accompanied by lengthening of the interval between uterine contractions and a shortening of the contraction time, it is assumed that the principal effect of the analgesic was that of reducing the intensity of the uterine activity. In all the patients studied there appears to be a close correlation between the pain intensity and length and frequency of uterine contractions.

Observations on the structure and function of the glomerulus in premature and full term infants. KENDRICK HARE, LUISE UNGEWITTER (by invitation), HENRY L. BARNETT (by invitation), and HELEN MCNAMARA (by invitation). *New York Hospital and the Departments of Pediatrics and Anatomy, Cornell University Medical College, New York City.* The anatomical development of the renal glomerulus has been followed in histological preparations from postmortem material from 29 fetuses and full term infants. The visceral layer of the epithelium is columnar in newly differentiated glomeruli and gradually passes through a cuboidal to a squamous form. Special silver staining reveals this layer to be a continuous sheet of extremely thin cells which contain a reticulum of argyrophilic fibrils. This process of differentiation is coincidental with the transition from the columnar to the squamous type of epithelium. It begins in the medullary layer of glomeruli and progresses toward the cortex to reach completion within a few months after birth. Rates of glomerular filtration were measured by inulin clearances in newborn premature and full term infants and at intervals thereafter until 6 months of age. These rates, low at birth, increased with age. The formation of a fine reticulated investment of the glomerular capillaried may be causally related to the increase in glomerular filtration.

Factors responsible for transmission of visible light by the fibrous tunic of the eye. WILLIAM M. HART and BRUCE F. CHANDLER (by invitation). *Depts. of Ophthalmology and Biochemistry, Temple University School of Medicine, Philadelphia.* In early embryologic development, the sclera of the eye is transparent. This fact must be accounted for in any attempt to explain the transparency of the cornea in the definitive eye. Studies on hydration properties of the cornea have shown no relation between the extent of swelling and scattering of light. However, there are differences in swelling behavior between cornea and sclera which can be attributed to the peculiar structural arrangement of the collagenous fibers in the two tissues. It is a well known observation that pressure on the intact eye produces a clouding of the cornea which disappears immediately as the pressure is released. This phenomenon is a true birefringence. The chief factor concerned in transparency of the cornea is the presence of this birefringent system which is due to the orderly arrangement of the corneal lamellae. The sclera became opaque in fetal development as a result of its closely felted structure which distorts the optic axes of its fibers. Drying causes corneal opacities to disappear. When such corneas are rehydrated the opacities do not reappear. This effect is due to rearrangement of the distorted corneal fibers. The translucency of sclera on dehydration has the same significance as the effect of drying on corneal opacity.

Retinal action potentials of photoreceptor cells and the discharge of nerve impulses in their axones. H. K. HARTLINE. *Johnson Research Foundation, University of Pennsylvania, Philadelphia.* Single ommatidia, containing 10–15 retinula cells with ½mm. of their attached axones, were isolated in the compound eye of *Limulus*. An oscillograph and D.C. amplifier were used to record the slow changes in electrical potential between one electrode placed on the piece of cornea to which the distal ends of the retinula cells were attached and another electrode on the proximal end of the ommatidium where the nerve strand emerge (retinal action potential). Simultaneously, impulses in the nerve strand were recorded by another amplifier and oscillograph. Upon illumination, the corneal electrode became more negative relative to the proximal electrode; at the same time impulses were discharged in the nerve strand. There was a rough parallelism between the rise and fall of the retinal action potential and the frequency of discharge of impulses; different intensities of stimulation and different conditions of adaptation had comparable effects on both responses. Closer analysis, however, showed that the

parallelisms were far from exact; in many preparations it was possible to elicit large changes in the impulse discharge with only very slight concomitant retinal action potentials. It is not yet possible to decide whether the local currents accompanying the retinal action potential may be considered the direct cause of the discharge of nerve impulses by the photoreceptor cells.

The utilization of carbohydrates by drosophila melanogaster. C. C. HASSETT (introduced by L. E. Chadwick). *Medical Division, Army Chemical Corps, Army Chemical Center, Maryland.* Drosophila can survive for varying periods on pure solutions of single substances including: pentoses, hexoses, di- and trisaccharides and the simpler polysaccharides, as well as on certain carbohydrate intermediates and other substances such as glycerol.

In vitro action of adrenal cortical extract upon lymphocytes. OSCAR HECHTER and DAVID STONE (by invitation). *Worcester Foundation for Experimental Biology, Shrewsbury, and Dept. of Physiology, Tufts Medical School.* It is now well established that adrenal cortical hormones (ACH) increase the rate of lymphocyte dissolution *in vivo*, principally within lymphoid structures (White and Dougherty, Ann. N. Y. Acad. Sci. **46**: 859, 1946). In previous work we have shown that addition of adrenal cortical extract (ACE) to blood has no direct effect upon blood lymphocytes; when, however, ACE was added to blood perfused through an isolated rabbit spleen, indirect evidence for accelerated lymphocyte breakdown was obtained, (Hechter and Stone, Endocrinology, in press). This finding suggested that the reaction of ACH to induce lymphocyte breakdown requires certain factors present in lymphoid tissue. In the present study, results will be presented which tend to show that the addition of ACE to breis or homogenates of rabbit lymphoid tissue, which are incubated with blood, accelerates the rate of lymphocyte breakdown, as evaluated by lymphocyte counts.

The effect of adrenocorticotropin on renal hemodynamics and uric acid clearance. LEON HELLMAN (by invitation), RAYMOND E. WESTON, DORIS J. W. ESCHER (by invitation) and LOUIS LEITER (by invitation). *Medical Division, Montefiore Hospital, New York 67, N. Y.* Purified adrenocorticotropin (Armour) was given by intramuscular injection (20 mg. every four hours for ten doses) to four patients. Glomerular filtration rate, renal plasma flow, urea, uric acid and creatinine clearances, and sodium and chloride excretons were determined before and after each series of injections. All patients had reduced GFR and RPF initially. In two patients, one with proved and the other with suspected hypopituitarism, there was a marked increase in GFR, RPF, and uric acid clearance after ACTH. In a patient with lymphatic leukemia, there was a marked increase in uric acid clearance and a significant, moderate increase in GFR and RPF. No change in GFR, RPF or uric acid clearance occurred in a case of pernicious anemia in relapse. Testosterone given to the patient with proved hypopituitarism caused only slight changes in renal function. All patients showed decrease in circulating eosinophils and/or lymphocytes after receiving ACTH.

Further studies on the identity of the sustained pressor principle. O. M. HELMER, ROBERT E. SHIPLEY and K. G. KOHLSTAEDT. *Lilly Laboratory for Clinical Research, Indianapolis General Hospital.* Intramuscular or intraperitoneal injection of various kidney extracts in several animal species causes the appearance in the sera of a substance or substances which are capable of destroying or "neutralizing" renin and the sustained pressor principle (Shipley et al., Am. J. Physiol. **149**: 708, 1947). In nephrectomized cats the intravenous injection of the antisera will lower the arterial pressure which has been previously elevated by the injection of plasma containing the sustained pressor material. It has not been determined whether or not the substance(s) in antisera which neutralizes renin is also responsible for the neutralization of the sustained pressor principle. The antirenin titers of the antisera have been determined by assaying the quantity of unneutralized renin. The assay is based upon the production of angiotonin.

Chronic hyperventilation in normal human subjects. A. HEMINGWAY, E. B. BROWN, G. S. CAMPBELL, F. GOLLAN and J. O. ELAM. *Department of Physiology, University of Minnesota.* Normal young adult male subjects were hyperventilated in a Drinker respirator for periods of 8 to 24 hours at ventilation ratios of 2 to 3. The ventilation was set at a value just below the threshhold for hyperventilation tetany. Analyses of the blood were made for CO_2 content and capacity, pH, acid-soluble and inorganic phosphorous, as well as in some experiments, sodium and chloride. Respiratory volumes and rates were measured before, during and after the experiment. Weakness, fatigue, headache and abdominal pains were caused by the long-continued hyperventilation. The usual respiratory alkalosis, denoted by lowered CO_2 content and elevated blood pH, was noted but the most striking feature was a reduction of the serum and urine inorganic phosphorous. These results have an obvious bearing on the management of respiratory patients.

The effect of blood acid-base changes on convulsive seizures. CHARLES D. HENDLEY (introduced by Horace W. Davenport). *Departments of Physiology and Pharmacology, University of Utah School of Medicine, Salt Lake City, Utah.* Metabolic acidosis or alkalosis was produced in rats by

feeding ammonium chloride, sodium bicarbonate, or sodium acetate. Respiratory acidosis was produced by administration of 10 per cent carbon dioxide in oxygen. Blood obtained by heart puncture was analyzed for pH and total carbon dioxide, and pCO_2 and extra fixed acid or base were calculated. Electroshock thresholds were determined, and the components of maximal seizure patterns were timed. Extreme metabolic acidosis (fixed acid = 24 ± 6 mM/l., pH = 6.81–7.06) raised the threshold 13 per cent (P = 0.01), but the same degree of acidosis (fixed acid = 23 ± 2 mM/l.) had no significant effect on seizure pattern. Mild metabolic acidosis (fixed acid = 6 ± 1 mM/l., pH = 7.25–7.40), mild or severe metabolic alkalosis (fixed base = 7 ± 2 mM/l., pH = 7.42 – 7.57 and fixed base = 15 ± 1 mM/l., pH = 7.53 – 7.61) and moderate to severe respiratory acidosis (pCO_2 = 48 – 101 mm. Hg, pH = 7.01 – 7.34) had no significant effect on threshold or seizure pattern (P is greater than 0.25). Moderate to severe metabolic acidosis or alkalosis in rabbits produced no significant effect on the resting EEG, on the subconvulsive Metrazol EEG, or on transcortical potentials evoked by condenser discharges. It is concluded that acute changes in blood acid-base balance must be very severe in order to cause any changes in the measures of cortical excitability used in these studies.

Tactile localization in the thalamus of cat and monkey. Elwood Henneman and Vernon Mountcastle (introduced by Philip Bard). *Dept. of Physiology, Johns Hopkins University, School of Medicine, Baltimore 5, Md.* The evoked potential method has been used to study the localization of tactile sensibility in the thalamus. The potential changes produced there by discrete tactile stimulation of the body surface were picked up by a monopolar electrode, carried by a stereotaxic apparatus. These potential changes were amplified and introduced into a loud speaker. By this method the area of skin whose stimulation produced potential changes at any one point within the thalamus was outlined. The point of maximal response within this area was determined oscillographically. The brains were sectioned serially, and the sections used to construct a three-dimensional model of the thalamus, within which the course of the needle tracts was determined. The responsive area in the thalamus of the cat is confined solely to pars externa and the pars arcuata of the ventro-lateral nucleus. Within these nuclei the volume of thalamic tissue devoted to a particular cutaneous area varies directly with the peripheral innervation density. The contralateral body surface is represented in great detail, with face postero-medially, and tail antero-laterally. The axial portions of the body from nose to tail are represented in continuity across the superior portion of the responsive area. There is, thus, no reversal of the cervical segments in the thalamus. The extremities are represented inferiorly. Ipsilateral face and mouth are represented medially to their contralateral locations. No other ipsilateral responses were noted.

The study has been extended to the Macaca mulatta.

The effect of pressure breathing on circulating blood volume. (1) Fluid loss into the tissue spaces. J. P. Henry, H. Jacobs (by invitation), H. Meeham (by invitation) and A. Karstens (by invitation). *Dept. of Physiology, University of Southern California, Los Angeles, California, and Aero Medical Laby., Air Materiel Command, Wright Field, Dayton, Ohio.* A constant elevation of the intrathoracic pressure of 20 to 60 mm. Hg results in a corresponding increase in mean venous pressure throughout the body. This in turn leads to a disturbance of the normal fluid equilibrium between blood and tissue fluid with significant hemoconcentration. Hematacrit and hemoglobin estimations were made on venous blood from eight subjects submitted to pressure breathing for 30 minutes at various pressures. In all cases there was hemo-concentration which increased in proportion to the pressure employed. At 30 mm. Hg mean intrathoracic pressure the fluid loss in 30 minutes in eight experiments was 4.0 ± 0.5 cc./100 cc. blood and at 45 mm. Hg in twenty experiments this loss increased to approximately 10.3 ± 0.5 cc./100 cc. blood. It is concluded that significant hemoconcentration is induced by breathing at pressures in the range of 30 to 50 mm. Hg for periods of 15 to 30 minutes.

The effect of caloric level of refeeding on the recovery of work capacity following semi-starvation. Austin Henschel, Henry Longstreet Taylor and Ancel Keys. *Laboratory of Physiological Hygiene, University of Minnesota, Minneapolis.* The effect of caloric intake level on rate of recovery of work capacity following semi-starvation was determined on 32 young men who lost 24 per cent of body weight in 6 months on a semi-starvation diet. Work capacity was measured by the Harvard Fitness Test. The average pre-starvation control Harvard Fitness Test score was 64.1 ± 17.2, time of run 242 ± 63.0 seconds and work accomplished 4501 ± 1173 kg. meters. Values at the end of semi-starvation, as per cent of control, were 28.4, 20.9 and 15.8 for the score, time of run and work accomplished, respectively. For 12 weeks of restricted rehabilitation the 32 men were divided into 4 groups of 8 men; the daily caloric intake of the groups differed progressively by about 350 Cal. with the weighted average caloric intake for this period being 2440, 2795, 3160, and 3495 for groups Z, L, G, and T, respectively. The fitness score at 6 weeks of rehabilitation, as per

cent of control score, was 31.2, 33.3, 48.6 and 49.6 for groups Z, L, G, and T, respectively, and 43.2, 50.9, 64.6 and 63.0 at 12 weeks. Substantially the same inter-group relationships held for time of run and total work accomplished. Recovery was significantly slower in groups Z and L than in the higher caloric groups but there was no significant difference between groups G and T. Supplementation of half the subjects in each group with extra vitamins had no beneficial effect on the rate of rehabilitation.

The effect of hyperthermia on the flow of thoracic duct lymph. J. F. HERRICK and J. L. BOLLMAN. *Division of Experimental Medicine, The Mayo Foundation.* Thoracic duct lymph was collected continuously from a small polyvinyl tube as a cannula in the thoracic duct. On the second or third day after operation the rate of flow was constant over a period of several hours when the unanesthetized rat was maintained at normal body temperature. The rate of flow was from 1 to 2 cc. each hour in the rats used. The rectal temperature was continuously recorded by means of a thermistor. Hyperthermia was induced by placing the rat in the radiation field of a microwave generator, the magnetron tube oscillating at a frequency of 2450 megacycles per second. The body temperature of the rat began to rise immediately and could be maintained at any desired level by altering the output of the generator. In most experiments when the body temperature was elevated more than two degrees C. in 15 minutes the flow of lymph was reduced for 15–30 minutes subsequently and then returned to the control levels. The return toward control levels occurred whether the hyperthermia was maintained or the animal allowed to return to normal body temperature unless lethal temperatures were reached. Hyperthermia was also produced by increasing the environmental temperature and humidity. The flow of lymph was similar to that observed with microwave induced hyperthermia.

Further studies on the correlation between skin volume pulses and blood flow. A. B. HERTZMAN and W. C. RANDALL. *Department of Physiology, St. Louis Univ. Medical School.* The correlation between the photoelectrically recorded skin pulse and the skin blood flow as demonstrated previously on the finger has been studied in other skin regions in the following experiments: (1) total basal cutaneous blood flow as calculated from thermal data were compared with the average skin pulse which in turn was calculated from a representative sampling of skin pulses from the various areas of the body. (2) Presumably maximal cutaneous blood flows in the forearm during histamine or mecholyl iontophoresis as calculated from the venous occlusion data were compared with the maximal amplitude of the skin pulses in the dilated areas. The two groups of data suggest that the flow equivalents of the cutaneous volume pulses as derived from previous calibration data on the finger are too large to be transferable to other skin regions. The lower flow equivalents in the latter may be due to differences in the vascular architecture since they are obtained at high and low flows. Fair uniformity in basal, resting and maximal flows is observed in the skin of the legs, arms, and trunk. This suggests uniform vasomotor tone and an approximately equivalent vascular bed in those areas. Higher relative and absolute values for flows in the hands, feet and face under similar conditions may be due to a combination of vasomotor and morphological factors.

Effects of oral cinchona alkaloids on circulation of dogs with experimental hypertension. EDWIN P. HIATT. *Department of Physiology, University of North Carolina School of Medicine, Chapel Hill, North Carolina.* Oral doses of quinine sulfate and quinidine sulfate of the order of 15 mg./kg. were given to dogs with experimental neurogenic hypertension. The effect of these drugs on the mean arterial blood pressure was determined by arterial puncture and a mercury manometer. The effective renal plasma flow and glomerular filtration rate were measured by p-amino hippurate and creatinine clearances. These alkaloids cause a marked fall in arterial blood pressure with a renal vasodilatation. The accelerated pulse rate is little affected. Normal control dogs show minor changes in blood pressure but a renal hyperemia and an increase in pulse rate. The effect of a single dose lasts 2–3 hours, but 4 doses at four hour intervals will cause the action to persist overnight. It can be maintained subsequently by two or three doses a day. During a course of such treatment the dogs continue to eat and behave normally. When the drug is discontinued after several days of administration the circulatory effects disappear in about 36 hours. The effective plasma concentrations of the alkaloids fall in the range of 1 to 3 mg./liter. In this range there is rough correlation between concentration and effect. Similar experiments are being carried out on dogs with experimental renal hypertension.

Is heat death due to a myotoxic factor? WILLIAM A. HIESTAND and FORST D. FULLER (by invitation). *Laboratory of Animal Physiology, Purdue University, Lafayette, Indiana and Dept. of Zoology, DePauw University, Greencastle, Indiana.* It has been reported (Fed. Proc. 5(1)43, 1946) that rats killed by immersion of the feet in water of 45°C. produce some substance(s) which if extracted from the skeletal muscles and injected into recipient rats will cause death in the latter. Our results do not corroborate these findings and

are stated here. Aqueous extracts of skeletal muscles of mice and rats exposed to heat were compared to similar extracts of muscles of control animals not exposed to heat and found to be no more toxic. Dialysates of such aqueous extracts when concentrated and injected into recipient mice cause a progression of symptoms about as follows: (1) opisthotonus of lower cord, (2) locomotor ataxia, (3) respiratory distress (panting, gasping, hiccoughing), (4) asthenia, (5) head tremors, (6) marked gasping, (7) convulsions, (8) death. It appears that part of the effects are due to stimulation of the central neuraxis beginning at the lower cord and progressing anteriorly until the animal dies of medullary paralysis. Statistical evaluation shows that no real difference exists between muscle extracts of heat-killed and non-heat-killed animals. The toxic symptoms are not due to intravascular thrombosis but resemble peptone injection. No increase was found in toxicity of the muscle extract in animals subjected to or killed by barrel "shock", suffocation, anoxia, as well as heat. Thus animals killed by exposure to heat appear to die from some cause other than a chemical substance toxic to the organism.

Di-isopropyl fluorophosphate (DFP): site of injection and variation in response. H. E. HIMWICH, and A. M. FREEDMAN (by invitation). *Army Chemical Center, Medical Division, Edgewood, Maryland.* Adult male albino rabbits under light pentothal anesthesia were injected into the femoral artery, femoral vein, carotid artery and portal vein with 0.4–0.8 mg./kgm. DFP in saline at rate of 0.01 mg./sec. In general, the most resistant animals were those injected into the portal vein. Even with the very high dosage of 0.8 mg./kgm. nicotinic and muscarinic effects were transitory. These animals made a prompt and uneventful recovery. The most sensitive group were those rabbits injected into the femoral vein. They displayed all the nicotinic and muscarinic effects of DFP toxicity and suffered a higher mortality than those rabbits injected into the femoral artery. The latter group when exposed to the lower dosage displayed only fibrillations in the leg supplied by the injected artery. In the higher dose range, systemic signs although marked were in each case less severe than in rabbits injected into the femoral vein with the same dose of DFP. The mortality of those animals injected into the carotid artery was similar to the femoral vein group. In all instances a marked and persistent unilateral meiosis was observed on the injected side. Half of the rabbits injected with 0.4 mg./kgm. in one carotid tipped their heads with their noses away from the side injected and made circular movements pivoting about on their hind limbs in a direction away from the injected side. The mechanism of these effects will be discussed.

Estimation of subcutaneous pressure in animals explosively decompressed to pressures of 30 mm. Hg. F. A. Hitchcock, Floyd M. Beman (by invitation) and JOHN A. KEMPH, (by invitation). *The Laboratory of Aviation Physiology, The Ohio State University, Columbus.* It has been noted that following explosive decompression of dogs to levels of approximately 30 mm. Hg a marked subcutaneous swelling develops. It has been postulated that this is due to the vaporization of body fluids. During other experiments, it has been noted that following explosive decompression there is a marked rise in venous pressure. It is thought that this rise in pressure may be due to an increase in the subcutaneous pressure. Arterial pressure is maintained at from 50 to 70 mm. Hg even though arterial pulse curves indicate that cardiac output ceases following explosive decompression. In preliminary experiments an attempt has been made to measure the subcutaneous pressure which develops during this swelling. Such determinations have been made both with the mercury manometer and with the electric capacitance manometer of Lilley. Results indicate that subcutaneous pressure following explosive decompression rises to a point between 29 and 34 mm. Hg when the ambient pressure is about 30 mm. Hg. Such pressures would be sufficient to prevent the vaporization of body fluids and it is believed that they are at least in part due to technical difficulties, perhaps the introduction of air under the skin at the time the needle is inserted. Crude determinations of the compositions of the subcutaneous gas indicate that it is chiefly water vapor.

Brain and muscle potassium in relation to stressful activities and adrenal cortical function. HUDSON HOAGLAND and DAVID STONE (by invitation). *Worcester Foundation for Experimental Biology, Shrewsbury, Mass.* Studies of the concentration of brain and muscle potassium of 300 rats were made under a variety of experimental conditions. Small (0.5 mg. per day) doses of desoxycorticosterone or of pregnenolone did not affect brain potassium in normal rats; desoxycorticosterone, but not pregnenolone, reduced the concentration of muscle potassium by 6.2%. Uncompensated adrenalectomy increased the concentration of brain potassium by 24.0% and of muscle potassium by 37.5%. Pregnenolone (0.5 mg. per day) lessened the increase in concentration of brain potassium in adrenalectomized rats by 10.0% and of muscle potassium concentration by 12.0%. The stress of prolonged swimming reduced the concentration of brain potassium by 4.4% and that of muscle by 6.1%. These together with the foregoing results, were all statistically significant ($P<0.01$). There was a suggestion that doses of 0.5 mg. per day of

both pregnenolone and desoxycorticosterone tended to prevent decrease in concentration of brain potassium due to prolonged swimming although the results were not statistically significant (P approximately 0.1). Pregnenolone also tended to prevent the decrease of potassium concentration in swim-stressed muscle but the P value was not significant (0.1 to 0.2). Desoxycorticosterone significantly augmented the decrease of concentration of muscle potassium in swim-stressed rats. Pregnenolone has been previously found by Pincus and Hoagland to enhance the efficiency of prolonged fatiguing psychomotor performance in man. Our present findings indicate that this substance exerts a homeostatic influence in the regulation of brain and muscle potassium.

Conduction velocity of skeleto-motor nerve fibers to partially paralyzed muscles in human poliomyelitis. ROBERT HODES. *From the Department of Physical Medicine, Graduate School of Medicine, and the Eldridge Reeves Johnson Foundation, University of Pennsylvania.* Maximal muscle action potentials, evoked by percutaneous stimulation of motor nerves and recorded by surface electrodes, were amplified by a condenser-coupled differential amplifier and photographed from a cathode-ray oscillograph. Conduction velocity of the most rapidly conducting nerve fibers supplying the intrinsic muscles of the hand and foot was determined by stimulating the nerve at two sites, measuring the latencies of the muscular responses evoked by each shock, and dividing the latency-difference by the distance between the points of stimulation. Amplitudes of electromyograms from some 300 weakened and normal muscles, when correlated with clinical evaluations of muscle power from these same muscles, indicated that the degree of paralysis in poliomyelitis could be reliably estimated by comparing potentials from paretic muscles with those from corresponding normal muscles. A linear correlation was found between the conduction velocity of the fastest nerve fiber in the residual innervation of a weakened muscle and the maximal electromyogram obtained from that muscle. This proportionality existed in 70 experiments covering a wide range of conduction velocities (the lowest being 29 meters per second, or 48% of normal) and potential amplitudes (the smallest being 0.2 mV, or 2% of normal).

The close correspondence between conduction rate and degree of paralysis is best explained by selective viral destruction of motoneurons having axons of large diameter. Histopathological and other data militate against indiscriminate depression of axonal excitability as the cause of the slower-than-normal conduction velocity in nerve fibers supplying paretic muscles in poliomyelitis.

Muscle action potentials in human poliomyelitis before and after treatment by Billig's method. ROBERT HODES. *From the Department of Physical Medicine, Graduate School of Medicine, and the Eldridge Reeves Johnson Foundation, University of Pennsylvania, Philadelphia.* Twenty-two patients in the convalescent stage of poliomyelitis who had shown no change in muscle power for at least one year were selected for electromyographic study. Maximal muscle action potentials, led from surface electrodes, amplified by a condenser-coupled differential amplifier, and photographed from a cathode-ray oscillograph, were obtained by percutaneous motor nerve stimulation. Changes from control action potential amplitude were followed 0.9 to 17.8 months after treatment by the "closed manual neurotripsy" method of Billig. Electrical activity was never observed after operation in muscles which had not yielded action potentials before neurotripsy. In partially paralyzed muscles the average potential amplitude, up to 4 months post-operatively, was less than the control. Four to 8 months after treatment the potentials were on the average slightly greater than before operation. At intervals greater than 8 months, operated muscles produced potentials whose average amplitude was 22% greater than their respective control values. The increase in potential size after 8 months was statistically significant. An occasional muscle, examined 1–1½ years after neurotripsy, showed a slightly smaller potential than the control. Even in those rare cases indications were that continued recovery would eventually produce muscle power equal to or exceeding the pre-operative level. The neuromuscular block observed in these patients before treatment (Hodes, R.: Am. J. Med. Sci., **213**: 509, 1947: Federation Proc., **6**: 130, 1947) was either abolished or markedly reduced after operation.

The pH of gastric mucous secretion after equilibration in vitro with alveolar air. FRANKLIN HOLLANDER and FRANCES U. LAUBER (by invitation). *Gastroenterology Research Laboratory, The Mount Sinai Hospital, New York City.* It was previously found that mucous secretion, collected from dogs' Heidenhain pouches after topical stimulation with various agents, possesses pH's as high as 9.2. When these data were grouped according to stimulus, the group means increased with increasing potency of the stimulus, the potency being judged by viscosity of the secretion. These observations are characteristic of this secretion as it occurs inside the gastric cavity, but such "alkaline" material probably undergoes CO_2-loss between secretion by the cells and collection from the pouch. Therefore, specimens collected in response to emulsions of eugenol (5%) and mustard oil (1%) were equilibrated with human "alveolar" air, and the resulting pH's determined. Similar studies were made on blood plasma. Loss of CO_2, absorbed during equilibration, was minimized by layering

mineral oil over samples during pH measurement. The plasma specimens had initial pH's (following CO_2 escape) of 7.68–8.11, which dropped to 7.3–7.6 after equilibration. For eugenol-stimulated mucus, the pH's fell to 7.4–7.5; the initial values being 7.9–8.6. For mustard oil secretion, the values dropped from 7.5–7.9 to 7.3–7.4. These results indicate that the pH of gastric mucous secretion after equilibration with "alveolar" air is constant and approximately the same as that of blood plasma. This suggests that the pH of gastric mucus as it exists inside the cell and at the instant of ejection into the gastric lumen is approximately that of venous blood.

Autotomy as a test for toxic factor A. L. HOPKINS (introduced by L. V. Heilbrunn). *Marine Biological Laboratory, Woods Hole, Mass.* When the leg of a crab is crushed, the animal typically responds by severing the injured appendage. This is the well known phenomenon of autotomy. It is logical to assume that one of the primary stimuli to autotomy is the production of an injury substance (toxic factor) by the injured muscles of the leg. Evidence for this point of view is given by experiments in which injury substances from frog muscles were injected into legs of the crabs Libinia emarginata and Sesarma cinereum. Injection of sea-water into such legs caused no autotomy. Nor did the injection of KCl cause autotomy in Sesarma. Also in Libinia preliminary results indicate that the legs do not drop off after KCl injection. On the other hand, injection of extracts of frog muscle caused as much as 50% autotomy of the injected legs in Sesarma and 30% in Libinia. The method of extraction makes it very improbable that the extracts contained any acetylcholine (which is known to cause autotomy). These experiments suggest that the autotomy of crab legs may be used as a means of testing for the presence of injury substances.

Standardization of a method for the production of experimental atherosclerosis in the chicken. L. HORLICK (by invitation) and L. N. KATZ. *Cardiovascular Department, Research Institute, Michael Reese Hospital, Chicago, Illinois.* The effect of varying concentrations of dietary cholesterol on the degree of hypercholesterolemia, the time of development of atheromatous lesions, and their severity were determined. Cholesterol feeding was instituted in four week old Leghorn cockerels. The results are given in the table herewith.

Dietary cholesterol	Per cent of animals in each group which developed atheroma at intervals indicated		
	At five weeks	Ten weeks	Fifteen weeks
Control	0	0	0
½%	25	80	66
1%	66	100	92
2%	75	100	92
4%	100	100	100

Feeding of cholesterol in the dosages employed produced a marked rise in the blood cholesterol in 6–12 times the normal levels within one week. There was a relationship between the amount of dietary cholesterol and the degree of the hypercholesterolemia. The critical dose of dietary cholesterol for cholesterolemia is between ½ and 1%. Doses above this do not produce any greater degree of hypercholesterolemia. The earliest atheromatous lesions occurred within two weeks after the commencement of cholesterol feeding. The frequency of occurrence and severity of the lesions varied directly with the amount of cholesterol in the diet and the duration of the feeding. In any individual group the severity of the lesions closely paralleled the degree and duration of the hypercholesterolemia.

Orthostatic hypotension following hot or cold baths. STEVEN M. HORVATH. *From Department of Physical Medicine, Graduate School of Medicine, University of Pennsylvania.* Transitory non-fatal collapse following severe physical effort is a familiar phenomenon in competitive sport. Similar syncopal responses are also observed in individuals given hot baths for therapeusis. However, observations dealing with the nature of this postural hypotension are still few. Subjects were immersed for a period of 20 minutes in a bath of either 18° or 40°C. Heart rate, blood pressure, cardiac output and body temperature measurements were made in the supine and erect (70°) positions before and after the bath. Position was always changed passively by means of a tilting ballistocardiograph. Approximately one-half of the subjects showed evidences of orthostatic hypotension following the hot bath. No subject showed such tendencies following the cold. The alterations in cardiovascular responses to changes in environmental temperature and posture will be discussed.

Direct experiments on the relation between the form of the ballistocardiogram and the shape of the systolic velocity curve in the aorta of man. ORVILLE HORWITZ (by invitation), ROBERT L. MAYOCK (by invitation), and ISAAC STARR. *From the Department of Therapeutic Research and the Department of Medicine, University of Pennsylvania.* As the first step of necropsy the aorta was cannulated through the aortic valve by a cannula of 2 cm. diameter, bent into a right angle and leading to a piston syringe so altered that the diameter of its outlet equalled that of the cannula. The cadaver was laid on the ballistocardiogram with the catheter in place. A reservoir filled with saline, placed 1.3 meters above the subject, was connected to a cannula in the femoral artery to keep the aorta full and provide a more normal pressure. An optical record of the piston's position at each instant

of time was recorded on the same film with the ballistocardiogram. A large variety of "systolic" ejection curves was secured by pushing or striking the piston in various manners. We have over 40 such experiments in 4 cadavers. When the ejection velocity curve was smooth and reached maximum velocity early in "systole" the ballistic form resembled that found in healthy persons during life, except that there was no H wave and I was smaller in relation to J than usual. When maximum velocity was attained late, or when the curve was not smooth, the ballistic form was highly abnormal. So the form of the ballistocardiogram was completely dependent on the shape of the aortic velocity curve and the results conformed in general to our expectations from theoretical considerations.

The knowledge gained will permit a more exact interpretation of abnormal ballistic forms in physiological terms.

Heating of human muscle tissue by micro waves. S. M. HORVATH, R. N. MILLER (by invitation) and B. K. HUTT (by invitation). *From the Department of Physical Medicine, Graduate School of Medicine, University of Pennsylvania.* The present modes of local heating are extremely inefficient in respect to their ability to penetrate deeply into tissues. It has been shown, however, that the employment of high frequency electrical energy can heat deeply and presumably quickly (although this latter point has defied experimental proof). While short wave diathermy (waves of several meters) is not readily adaptable to the heating of small areas, recent availability of generators producing wave lengths in the centimeter range which can be focused on small areas offers a tool which could provide such localization. The evaluation of Micro Wave energy sources as a means of providing deep and localized heating of tissues was made on eight subjects. These waves were directed at the outer lateral surface of the thigh for a period of fifteen minutes with the power loads varying in different experiments from 32 to 112 watts. Surface, subcutaneous and muscle (1 to 2 inches) temperatures were measured prior to and immediately following the application of the heat source. Observations were continued for periods of one to two hours. Due to the properties of the waves it was not feasible to measure temperatures with thermocouples during the exposure. In general the increase in temperature in the surface, subcutaneous and muscular tissues was roughly equivalent, a matter of 3° to 4°C. In a number of instances subcutaneous temperatures were appreciably higher than elsewhere in the area, and values as high as 44°C. were recorded. All temperature changes were definitely localized in the sense that no increases were observed outside the area immediately under the directors employed to project the high frequency waves. As far as is known at present, their maximum depth of penetration appears to be approximately two inches. Muscle temperatures of the opposite side of the thigh did not differ appreciably from control values. No changes in body (rectal) temperature were observed even when exposure periods were lengthened to thirty minutes.

Statistical analysis of filtration rate and renal plasma flow in normal dog and man. C. RILEY HOUCK. *From the Departments of Physiology, New York University College of Medicine, New York and the University of Tennessee College of Medicine, Memphis, Tennessee.* The filtration rate (creatinine clearance) and the effective renal plasma flow (para-aminohippuric acid clearance) in 75 normal, trained, fasted, conscious female dogs have been analyzed. Related to surface area, filtration rate has a mean of 84.4 cc./min./m²; range, 43–133; standard deviation, 19.1; coefficient of variation (100 × standard deviation/mean), 22.6. Plasma flow has a mean of 266 cc./min./m²; range, 139–430; standard deviation, 66; coefficient of variation, 24.8. The coefficient of correlation (r) between these functions is 0.79. Related to body weight, filtration rate has a mean of 4.29 cc./min./kg; range, 2.15–8.32; standard deviation, 1.01; coefficient of variation, 23.6. Plasma flow has a mean of 13.51 cc./min./kg; range, 8.05–22.43; standard deviation, 3.26; coefficient of variation, 24.1. The coefficient of correlation is 0.73. In a comparable analysis in 61 normal men (data from H. W. Smith, Lectures on the Kidney, University of Kansas, Lawrence, 1943), filtration rate has a mean of 75 cc./min./m²; range, 51.4–103.2; standard deviation, 12.7; coefficient of variation, 16.9. Plasma flow has a mean of 403 cc./min./m²; range, 198–546; standard deviation, 78.5; coefficient of variation, 19.5. The coefficient of correlation between these functions is 0.82. The dog has a greater filtration rate than man on a surface area basis, but a smaller effective renal plasma flow. Therefore, the filtration fraction (filtration/flow) in the dog (0.317) is higher than in man (0.186). The correlation between filtration and flow seems only slightly better in man than in the dog.

Chemical studies of sex steroid balance in human subjects. F. D. HUMM (by invitation) and W. T. SALTER. *From the Laboratories of Pharmacology and Toxicology, Yale University School of Medicine, New Haven, Connecticut.* When the sex steroid hormones in the urine are evaluated for normal subjects, cancerous individuals and patients exhibiting various endocrinopathies, characteristic values are obtained by chemical methods. In the normal female, near the menstrual period the value for "estroid" excretion is frequently close to the normal male range; whereas

at the time of ovulation it rises to a sharp peak. In general, however, the range of "estroid" excretion as determined chemically in the urine of young females is definitely higher than the range for normal males. Similarly the range of 17-ketosteroid excretion is usually definitely below that for males. This discrepancy between male and female becomes accentuated if 1000 × the ratio of "estroid" to 17-ketosteroids is determined.

The values so obtained by chemical methods are at least as significant as biological assays. Chemical methods have the advantage of greater convenience and somewhat greater accuracy. Characteristic chemical values for "estroids" (micrograms per 24 hours) and 17-ketosteroids (milligrams per 24 hours) follow:

Normal woman (aged 25) at ovulation: 59 and 7.8
Adrenal virilism, female, aged 42: 44 and 42.9
Gynecomastia, male, aged 27: 8 and 7.9
Hypogonadism, male, aged 19: 4 and 6.1
Hypopituitarism, male, aged 47: 0 and 1.9
Fibroadenoma of breast, female, aged 26: 10 and 14.6
Breast carcinoma, female, aged 54: 4 and 3.4

Further detailed data in characteristic cases are discussed. In particular, the effect of the administration of stilbestrol and testosterone are noted.

Muscular atrophy as a state of "local hyperthyroidism". ERNST G. HUFF (introduced by Ernst Fischer). *Baruch Center of Physical Medicine and Department of Physiology, Medical College of Virginia, Richmond.* Several biochemical data suggested that the state of a denervated muscle can be regarded as a "local hyperthyroidism". Two possibilities were regarded: 1) The production of thyroxin in the thyroid gland has a greater effect on the denervated than on the normal muscle. 2) The extrathyroidal production of thyroxin in the denervated muscle is different from that in the normal muscle. Experiments on rats: Thyroxin (sub.cut.; 2 weeks) accelerates, while thiourea inhibits the atrophy. The differences were small, but particularly in the case of thyroxin highly significant. The effects cannot be explained merely by movement of water. The differences are also significant by calculating on the basis of the dried muscle. Arsenic and 2,4-Dinitrophenol accelerate the atrophy also. The degree of atrophy when completely carbohydrate-free or fat-free diets were given was the same as when mixed diet was given. Complete protein-free diet retards the atrophy. The differences were significant, for both wet and dry muscle weight. This could be the result of the lack of "specific dynamic action" of protein. Dose-response curves for thiourea: At all doses (2.5–40 mg./100 g.) the thyroid glands of control rats were slightly heavier than those of rats with both the ischiadic nerves cut. The diet of those rats was dog chow, which is rich in iodine. By giving a low iodine content diet (Gluten, Cornmeal, Corn Oil, $CaCo_3$, NaCl), the dose response curves show the opposite relations. At all doses, the thyroid glands of control rats were slightly lighter than those of rats with both ischiadic nerves cut.

Effects of tris-(β-chloroethyl) amine on respiration, carbohydrate and protein synthesis, and cell division in chilomonas. JOHN O. HUTCHENS, BETTY PODOLSKY (by invitation) and T. M. MCMAHON (by invitation). *Toxicity Laboratory and Department of Physiology, University of Chicago.* *Chilomonas paramecium* grows rapidly in pure culture in solutions containing carbon as acetate and nitrogen as ammonia. Effects of tris-(β-chloroethyl) amine (HN-3) on rate of oxygen consumption, utilization of acetate, utilization of ammonia, synthesis of starch, synthesis of protein, and rate of cell division in this organism have been studied. When HN-3 in concentrations of 10^{-6} to 5×10^{-4} M is added to cultures and the cells incubated in the presence of the HN-3 and/or its hydrolysis products the following effects are observed: (1) Oxygen consumption is not significantly depressed. (2) Acetate utilization and starch synthesis are not significantly depressed. (3) Ammonia utilization and protein synthesis are markedly inhibited. (4) Cell division is inhibited. The net effect is an increase in cell size, each cell containing a greater amount and proportion of starch and a smaller proportion of protein. At times varying from 5–15 hours, depending on HN-3 concentration, the rate of cell division returns toward normal and the cells tend toward normal size and content.

When 10^{-3} M HN-3 is employed, as above, death of the cells occurs within one hour. Brief exposures to this concentration followed by washing of the cells permits isolation of a small proportion of survivors. Pure clones so isolated show persisting attenuated rates of growth, respiration, and synthesis of components for at least several months.

Reflex activities and peristaltic function of the esophagus. KAO HWANG (introduced by A. C. Ivy). *Dept. of Clinical Science, University of Illinois College of Medicine, Chicago.* We have previously reported that the cervical portion of the esophagus in the dog receives from the vagus a separate motor nerve supply which has been designated as the pharyngo-esophageal nerve and was found to be responsible for all the motor activities of the cervical esophagus, peristaltic or of any other origin. When a gasping type of respiration developed in anesthetized dogs, the cervical portion or sometimes the entire length of the esophagus manifested spasmodic contractions synchronous with the inspiratory phase. The

cervical component of this contraction was abolished after bilateral section of the pharyngo-esophageal nerve. Stimulation of the central end of the vagus nerve is known to produce reflex contraction of mainly the cervical esophagus. This was found to be an ipsilateral reflex, being abolished by section of the pharyngo-esophageal nerve of the same side. Under morphine and procaine infiltration anesthesia the peristaltic function of the esophagus of the dog could be studied by introducing a balloon through the stoma of an esophagostomy located at the base of the neck. Secondary peristalsis was demonstrated throughout the entire length of the esophagus. A swallowing reflex was shown to arise from the cervical esophagus on distension. The pharyngo-esophageal nerve was found to be merely the efferent pathway for the peristaltic function of the cervical esophagus, both primary and secondary. Section of the recurrent laryngeal nerves close to the cricoid cartilage abolished in most dogs the swallowing reflex and also the secondary peristalsis of the cervical esophagus while the primary peristalsis was preserved.

Changes in urinary glucose and nitrogen following adrenalectomy in the force-fed diabetic rat. D. J. INGLE and M. C. PRESTRUD (by invitation). *Research Laboratories, The Upjohn Company, Kalamazoo, Michigan.* Earlier studies of adrenalectomy in diabetic animals indicated that the amelioration of the diabetes was due principally to a decrease in gluconeogenesis from protein, as evidenced by the striking decrease in urinary nitrogen. Consider: (1) the abnormally high food-intake of the diabetic animal is depressed to subnormal levels by adrenalectomy; (2) the abnormally high nitrogen loss of the diabetic animal falls to normal when the wasting of glucose is stopped. On these grounds it can be postulated that the change in nitrogen balance following adrenalectomy is secondary to an improvement in the utilization of carbohydrate and to changes in food-intake.

Partially depancreatized male rats of the Sprague-Dawley strain were force-fed a medium carbohydrate diet and given saline to drink throughout the experiment. They excreted an average of 3.5 to 4.0 grams of glucose per day. The nitrogen loss was greater than normal. Following adrenalectomy there was a sharp rise in nitrogen loss by the third day which coincided with a fall in urinary glucose. The urinary glucose disappeared and the urinary nitrogen fell gradually to normal values, but not below. A decrease in gluconeogenesis from protein, as evidenced by the change in urinary nitrogen, could account but for little glucose. Adrenalectomy either caused a change in gluconeogenesis from fat or the animals regained the ability to utilize (oxidation, storage or conversion) the carbohydrate of the diet which they wasted into the urine prior to adrenalectomy.

Further observations on the effect of pentobarbital and of an adrenolytic agent upon the survival of animals subjected to a procedure resulting in experimental hemorrhagic shock. R. C. INGRAHAM and FRANKLIN ROEMHILD (by invitation), and H. GOLDBERG (by invitation). *Department of Physiology, University of Illinois College of Medicine, Chicago.* We have previously observed that animals hemorrhaged to establish a blood pressure of 35–38 mm. Hg and maintained at this constant level for 90 min., pass into irreversible hemorrhagic shock in 70% of all cases and die despite transfusion. It was further observed that carefully designed therapy with pentobarbital and dibenamine had a marked beneficial effect on the survival of these animals. It was desired in these experiments to attempt to evaluate the efficiency of these agents upon the survival of animals in which the impending shock state is established by hemorrhaging the animal until the residual blood volume had been reduced below the generally accepted 60% of normal. In this type of procedure no attempt is made to maintain the blood pressure at any given constant level during the course of hypotension. Twenty dogs were subjected to the Wolcott method of producing the hemorrhagic shock state. It was intended that three hours after hemorrhage, the animals would be reinfused with all the remaining withdrawn blood. It was found 55% could not withstand a period of hypotension of this length. Fatality rate for the entire series was 80%. In the second series of 20 dogs, 7 mg./kg. of pentobarbital was injected slowly 10 min. after hemorrhage. Additional smaller amounts were injected as required to maintain the animals in a light state of sedation. 40% of the animals in this series were not able to withstand the three hour period of hypotension. The fatality rate for this series was 70%. In the third series, the adrenolytic compound was administered in doses of 3 to 1 mg./kg. 10 min. after hemorrhage. 44% of the animals in this series either died or showed definite signs of dying from cardio-respiratory before the three hour period had elapsed. 56% withstood the three hour period of drastic hypotension and the fatality rate for the entire series was 73%.

The effect of pressure breathing on circulating blood volume. (2) Distension of the venous reservoirs. H. JACOBS (by invitation), A. KARSTENS (by invitation) and J. P. HENRY. *Aero Medical Laby., Air Materiel Command, Wright Field, Dayton, Ohio.* In addition to inducing a loss of fluid from the circulation, an increase in venous pressure induced by pressure breathing distends the veins that lie outside the raised pressure area of the head and trunk thus pooling a certain por-

tion of the available circulating blood. The method of Ebert and Stead, J.C.L. **19**: 561, 1940, was applied to an estimation of the volume of blood sequestered from the circulation during pressure breathing. An approximate determination was made by this method of differences of the volume of blood in the two legs and one arm, isolated by application of arterial occlusion cuffs. The results of comparison made between the volumes thus obtained without and with pressure breathing at 40 mm. Hg for 2 minutes were variable. However, the approximate mean volume change due to blood sequestered in the legs and one arm in four subjects, each submitted to three series of estimations, was calculated at approximately 300 cc. or 6% of the total blood volume. This change is in fair agreement with estimations made by plethysmographic and teeter-board methods. When combined with the loss as determined from hemoconcentration studies, the estimated total loss of circulating blood volume when breathing for 30 minutes at 50 mm. Hg pressure amounts to as much as 15 to 20 per cent. Still higher pressures will lead to even greater losses of blood volume. These changes are in themselves adequate to account for the frequent development of syncope during periods of raised intrathoracic pressure.

The effect of parathormone on the renal tubular reabsorption of inorganic phosphate. IFTAKHAR JAHAN (Post-graduate fellow of the Govt. of India) (by invitation) and R. F. PITTS. *Department of Physiology, Syracuse University College of Medicine, Syracuse, New York.* The effect of parathormone on the renal tubular reabsorption of inorganic phosphate has been studied in twelve experiments on two female dogs. Six experiments were performed with and six without parathormone, the latter serving as controls. In the ones with parathormone the extract (Lilly) was given subcutaneously in two injections of 3 cc. (U.S.P. 300 units) each, separated by in interval of 6 hours, starting about 20 hours previous to the experiment. The plasma inorganic phosphate was varied over a range of 0.9 to 5.58 mM/liter by intravenous infusion of inorganic phosphate of pH 7.4. The rate of glomerular filtration was measured by creatinine clearance. Prior to the administration of phosphate there was no apparent difference either in the plasma level or in the rate of excretion of phosphate in the six control experiments and in the six in which parathormone was administered. In contrast, the plasma calcium level and the rate of calcium excretion were considerably elevated in the parathormone experiments in comparison with the control ones. When the plasma level of inorganic phosphate was increased from 0.9 to 5.58 mM/liter, the rate of renal tubular reabsorption of phosphate was the same in the parathormone treated animals and in the controls within limits of experimental error. It is concluded that increases in plasma calcium level and in the rate of calcium excretion in parathormone treated animals are not related to effects of parathormone on the phosphate reabsorptive mechanism of the kidney.

Pulmonary gas exchange following ligation of a pulmonary artery in man. O. H. JANTON (by invitation), H. P. REDONDO (by invitation) and J. C. SCOTT. *Department of Physiology, The Hahnemann Medical College, Philadelphia, Pennsylvania.* A 21 year old male at operation on 10/24/47 for ligation of a recanalized patent ductus arteriosus was found to have had a completely ligated left pulmonary artery (done in 1941) and a patent ductus. No gross pathology of the left lung was noted except for a residual adhesive pleuritis. The ductus was ligated and patient made an uneventful recovery. Angiography gave no evidence of left pulmonary circulation. On 11/18/47 samples of air were collected by a bronchospirometric catheter (Gebauer) simultaneously from the right and left sides at the end of a forced expiration following a normal expiration. Three pairs of samples were collected at intervals of 3 or 4 minutes. During procedure patient was uncomfortable and dyspneic. Average Co_2 and O_2 values for the left side were 3.74% and 19.14% respectively; for the right side 6.15% and 10.51%. Successive pairs of samples agreed approximately, although there was a progressive fall in oxygen on the right side. The difference in CO_2 and O_2 from room air, in samples obtained from the left side could have been due to mixing of the right and left lung air during periods of breathing between collection of samples as the two sides were sealed by inflatable cuffs only during the time of sampling. A bronchial artery anastomosis with pulmonary capillary bed may have accounted for CO_2, O_2 values on the left side but no evidence for this assumption is available. Normal tidal air was 879 cc. which supports the assumption that little gaseous exchange was occurring on left side.

Thalamo-cortical systems and the electrical activity of the brain. HERBERT H. JASPER and JAN DROOGLEEVER-FORTUYN (by invitation). *From the Department of Neurology and Neurosurgery of McGill University and the Montreal Neurological Institute.* Local electrical activity from specific cortical areas, and from certain subcortical structures was recorded in cats and monkeys during light barbiturate anesthesia, or in unanesthetized animals following medullary section as described by Bremer. The electrical response of the cortex to visual, auditory, and somato-sensory afferent impulses was characterized not only by evoked potentials in local primary (and secondary) receiving areas, but also by a less well localized

rhythmic response in the receiving area as a whole, secondary responses in many other cortical areas, and by a desynchronization of rhythmic activity in associated areas. Under certain conditions it seemed that a large portion of the cortex reacted in some way to a single specific afferent stimulus. Local stimulation of thalamic relay nuclei produced similar local effects in their areas of cortical projection with less generalized response. Control of rhythmic activity over widespread areas of the cortex was possible by local stimulation of the intralaminar nuclei of the thalamus and the n. reticularis. The distribution of patterns of spontaneous electrical activity seemed to follow units of nuclear thalamo-cortical projection systems. Integration and synchronization of these units was affected by the intralaminar and reticular systems of the thalamus. The pattern of rhythmic electrical activity characteristic of a given cortical area was abolished if its thalamic nucleus was destroyed.

Arterial pulses simulated in electrical analogues of the circulatory system. KENNETH E. JOCHIM. *Dept. of Physiology, Univ. of Kansas, Lawrence.* Previous reports (Fed. Proc. **1:** 43, 1942; **5:** 52, 1946; **6:**136, 1947) have dealt with arterial pressure pulses and flow pulses recorded from a simplified mechanical model of the circulatory system, and have presented a mathematical analysis of the behavior of such a model. It has been found possible to set up various types of pulsed electrical networks as analogues of the circulatory system. In these networks the circuit parameters of capacitance, resistance and inductance are analogous, respectively, to vascular distensibility, resistance to blood flow and mass of moving blood. Voltage and current pulses were recorded from these circuits and compared with corresponding pressure and flow pulses obtained on animals. Factors determing the wave form of the electrical pulses were analyzed and the possible influence of analogous factors on arterial pulses in the animal were indicated. The use of electrical circuit analogues in the study of the circulatory system has the following advantages: (1) with electrical networks it is relatively easy to simulate in the laboratory many of the complex characteristics of the circulation, (2) the various circuit parameters can be accurately controlled, and (3) precise measurements of all variables can be made.

Studies on deceleration. MILTON JOFFE (by invitation) and F. A. HITCHCOCK. *The Laboratory of Aviation Physiology, the Ohio State University, Columbus.* In order to investigate the physiological effects of rapid, severe deceleration it is first necessary to know the strength of the structural components affected and their resistance to deceleration forces. The literature on the subject is nil. Thoraces of normal human individuals were obtained and the physical properties of the ribs investigated by means of the Olsen Universal Lever Type Testing machine which applied an anterior posterior compression to the rib to the point of destruction. All ribs tested broke in the anterior third at an average value of about 15 lbs. Curves of load vs. distortion were drawn, and calculations of total energy absorbed were made. All ribs absorbed energy in the range of 10–100 inch pounds. Obviously the thorax can absorb more energy, up to 1000 foot lbs. or more, and we conclude that the bony structures contribute little to the total strength. Bone ash determinations made on fat free, dry bone showed no variation correlating with strength. The strength of the intact human thorax by means of simulated decelerations is being observed by means of the fluoroscope. Changes in the viscera are also observed and animal experiments to determine changes in cardiology (pressure and E.K.G.) with similar constriction of the thorax and abdomen are under way.

The thermal denaturation of tobacco mosaic virus in relation to hydrostatic pressure. F. H. JOHNSON, M. B. BAYLOR (by invitation) and D. FRASER (by invitation). *Department of Biology, Princeton University, Princeton, N. J.* The thermal denaturation of tobacco mosaic virus, as measured by decrease in soluble protein, at pH 7.0 in 0.1 M phosphate buffer, follows the kinetics of a first order reaction at both normal and increased hydrostatic pressures. Pressure retards the reaction, the rate constant changing from 0.00759 min.$^{-1}$ at 68.8°C. and normal pressure, to 0.00136 min.$^{-1}$ under 7,000 lbs. per sq. in., indicating a volume increase of activation, $\Delta V\ddagger$, of very close to 100 cc. per mole. Using this value for $\Delta V\ddagger$, and 153,000 calories for $\Delta H\ddagger$ as reported by Lauffer and Price (Jour. Biol. Chem., **133:** 1, 1940), rates calculated according to the Theory of Absolute Reaction Rates for various pressures up to 10,000 lbs. per sq. in., and 72.5° as well as 68.8°C., were found to agree closely with the data from experiments. The free energy of activation, $\Delta F\ddagger$, at normal pressure was computed to be 26,200 calories at 68.8° and 24,819 calories at 72.5°C., with an entropy of activation, $\Delta E\ddagger$, of 370.89 calories per degree. The $\Delta V\ddagger$ of 100 cc. per mole is of the same order of magnitude as that which has been reported for proteins of much lower molecular weight than tobacco mosaic virus, e. g., human serum globulin as well as intracellular and extracted enzymes.

Electrochemical determination of ionic diffusion through the synovial membrane. N. R. JOSEPH (by invitation), C. I. REED and I. E. STECK (by invitation). *Department of Physiology, College of Medicine and Department of Chemistry, College*

of Pharmacy, University of Illinois. Electrochemical methods of determining diffusion potentials have been applied *in vivo* to the study of the synovial membrane. The electrodes were the Ag-AgCl type in identical 0.15 m NaCl solution. The reference electrode was connected to the same solution inserted subcutaneously through an insulated needle outside the knee joint capsule of dogs. The indicator electrode was connected by means of a saturated KCl agar bridge to solution within the joint. Electrical contact was made through an insulated needle inserted into the joint cavity. Solutions of various electrolytes of any concentration were introduced into the joint and the differences of potential observed. In addition, currents through external resistance were determined, with or without applied external E.M.F. Polarization effects were also observed. For most inorganic ions of the alkali, alkali earth and halogen series, the resting potentials are stable and reversible. The relative ionic mobilities correspond in many cases to those in water. Iodide gives a high positive potential, indicating high mobility in the tissues, while calcium and magnesium ions were also quite positive, indicating low mobilities. Salicylate and benzoate showed lower mobility than chloride, the least diffusible of the halides. Their mobility relative to chloride was greater in tissues than in water. Calculations were based on the Planck-Henderson theory of diffusion potentials. Acetyl salicylate ion and its 5 brom derivative in dilute solutions made isotonic with glucose showed initially high positive potentials that rapidly drifted negative. Similar results were observed with CNS ion. Studies of currents through an external circuit give data on the stability of the diffusion potentials, on specific rates of transference of ions and on the resistance of the membrane.

Determination of circulation rate in articular structures. E. KAPLAN and N. R. JOSEPH (introduced by C. I. Reed). *Department of Physiology, College of Medicine and Department of Chemistry, College of Pharmacy, University of Illinois.* The experimental method is to increase the temperature within the knee joint of dogs and to determine the subsequent rate of cooling. Temperature is increased by electrically heating for one minute a needle inserted into the joint cavity. The needle is wound with a number of turns of fine insulated copper wire which conducts the heating current. This is regulated to raise the temperature of the joint about 5°C. After the heating period the temperature is allowed to equilibrate. Temperatures are observed by means of a thermocouple inserted through the needle. When the logarithm of temperature displacement is plotted against time of cooling, a linear relation is found: $\log \Delta T = \log \Delta T^\circ - kt$. where ΔT is the temperature displacement from equilibrium at time t, ΔT° is the initial displacement when t is zero, and k is a constant. Theoretically k is proportional to the circulation rate and inversely proportional to the volume of the static fluid which is cooled. The relation is of the form: $2.303\ k = (a + r/v)$, where r is the circulation rate in cc. per unit time, v is the volume of the static fluid in cc., and a is a constant proportional to the static cooling rate when r is zero. As an average for dogs, k is approximately 0.4 reciprocal minutes. Evaluation of the data shows that on the average a is 0.17 reciprocal minutes and r/v is 0.75. Thus the normal circulation rate in the articular structures is 0.75 cc. per minute per cc. of static fluid. Variations of rates under various experimental conditions such as external changes of temperature and administration of drugs have been observed.

Seismography applied to the study of track running. PETER V. KARPOVICH and NATHAN MILLMAN (by invitation). *Dept. of Physiology, Springfield College, Springfield, Mass.* A new method for the study of track running has been developed. It employs a number of seismometers placed along the track at 20 yard intervals and the direct measuring of the distances between the footprints left on lime sprinkled over the cinders. The impacts made by runners' feet on the track cause oscillations of the ground, which are picked up by the seismometers and are recorded on a portable electrocardiograph. (Sanborn Cardiette) The time intervals are also recorded in $\frac{1}{10}$ of a second. This method enables to calculate the following: a—velocity of running at each step; b—duration of each step; c—length of each step. A few preliminary tests indicated that in sprint running the step made with the right leg is usually greater in length and faster than that made with left leg. This is because most people are right handed and their *left legs* are stronger than their right legs. Left handed people will have the reverse. An exception was observed in one left-handed man who developed athletic habits of a right handed man and therefore had his left leg stronger than the right one. During distance runs no difference in the length of steps has been observed.

A method of determining the spread of excitation in the ventricles of the dog's heart. W. KAUFMAN (by invitation), H. M. CHERNOFF (by invitation) and L. H. NAHUM. *Laby. of Physiology, Yale Univ. School of Medicine, New Haven, Conn.* In any lead, during the inscription of the QRS complex, an upward deflection indicates electrical effects preponderantly from a distal area; a downward deflection indicates electrical effects preponderantly from a proximal area; and no deflection indicates that excitation is occurring either in an intermediate area, or simultaneously in a proximal and distal region where the electrical effects are

equal, but oppositely directed. The ventricular region being depolarized at any given time can be identified by an analysis of the positions of instantaneous points of the QRS complex in three simultaneously recorded unipolar limb leads (Vr, Vl, Vf). The sequence of ventricular excitation can be determined by the analysis, at successive time intervals, of instantaneous points of the QRS complex in the three simultaneously recorded unipolar limb leads. In dogs with intact chests, the first region of ventricular excitation lies within the mid-portion of the left anterior ventricle, near the septum; then, excitation spreads successively to the right anterior ventricle, to the posterior septum and posterior right ventricle, and to the apex and left posterior ventricle. The regions last depolarized lie in the right anterior ventricle near the pulmonary conus, and in the uppermost portion of the left posterior ventricle. These findings obtained by an indirect method are in accord with the results of direct measurements of the order of ventricular excitation of the dog's heart.

The effects of insulin hypoglycemia and coma on human cerebral metabolism and blood flow. SEYMOUR S. KETY, FRANCIS D. W. LUKENS, RACHEL B. WOODFORD (by invitation), MEREL H. HARMEL (by invitation), F. A. FREYHAN (by invitation), and CARL F. SCHMIDT. *Departments of Pharmacology, Medicine and Psychiatry, University of Pennsylvania and the Delaware State Hospital.* Measurements of cerebral blood flow by means of the nitrous oxide technique permitted determination of cerebral oxygen and glucose utilization in 5 schizophrenics in the resting state and in hypoglycemia both with and without coma. Average results are presented in the table. The arterial glucose shows a progressive fall while the oxygen and carbon dioxide contents are not significantly changed. The mean arterial blood pressure and the cerebral blood flow were not significantly affected. There is a progressive fall in both oxygen and blood glucose utilization by the brain as hypoglycemia and coma develop and these changes are statistically significant. The cerebral RQ is not significantly changed from unity throughout. The ratio of oxygen consumed to blood glucose utilized by the brain confirms the conclusion of earlier workers that glucose is the sole foodstuff of the resting brain. The increase in this ratio as hypoglycemia progresses indicates either the oxidation of some other substance or the utilization by the brain of its own carbohydrate stores.

	Control	Hypoglycemia without coma	Insulin coma
Arterial			
Glucose mg. %	74	19	8
CO_2 vol. %	52.1	50.6	50.8
O_2 vol. %	17.4	17.9	16.6
Mean blood pressure mm. Hg	94	86	93
Cerebral			
A-VO_2 vol. %	5.9	4.4	2.8
Blood flow cc./100 g./min.	58	61	63
O_2 consumption cc./100 g./min.	3.4	2.6	1.9
Glucose consumption mg./100 g./min.	4.4	2.3	0.8
R. Q.	0.95	1.10	0.92
O_2/glucose consumption	0.77	1.13	2.38

Tryptic activity of pancreatic juice after dilution with gastric juice. ELIZABETH N. KING (by invitation), M. H. F. FREIDMAN, and I. J. PINCUS (by invitation). *Department of Physiology, Jefferson Medical College, Philadelphia.* In tests of human pancreatic function gastric and duodenal contents are collected separately. However, constant aspiration applied to the stomach does not always prevent dilution of the duodenal contents with acid gastric chyme. In normal subjects following secretin administration the duodenal contents show a high initial rise in trypsin concentration: this does not occur in patients with proved chronic pancreatitis. The question arose whether in normal subjects the tryptic activity of secretin-stimulated pancreatic juice could be reduced to the low values seen in chronic pancreatitis by admixture of duodenal contents with gastric contents. Tryptic activity and pH of dog's pancreatic juice and human duodenal contents were determined before and after serial dilution with 0.1 N HCl, gastric juice, gastric juice boiled to inactivate pepsin, or water. Reduction of tryptic activity by 20% occurred when pancreatic juice was diluted with water to 45% of its original concentration. Tryptic activity was reduced 20% or more only when the pH was depressed below 6.0; this required dilution of the pancreatic juice to 70% with gastric juice, boiled gastric juice, or acid. Following secretin injection to 70 normal fasting subjects, 92 to 210 cc. duodenal contents and 1 to 38 cc. gastric contents were recovered in one hour. It is concluded that probably only in patients with gastric hypersecretion would enough acid gastric juice be secreted to inactivate sufficient trypsin to give the low values for tryptic activity shown by the duodenal contents of patients with chronic pancreatitis.

Effect of corticotropin on ovariectomized C3H mice bearing adrenal adenomas. JOSEPH T. KING, CARMEN B. CASAS (by invitation) and CLAIRE J. CARR (by invitation). *Department of Physiology, University of Minnesota.* Female mice of the C3H strain castrated at an early age are known to develop adrenal cortical adenomas and to come into constant subestrus on full feeding. We have previously reported that, when restricted in caloric intake to 66% or less, the ovariectomized

mouse still develops the adrenal tumor seen in the full-fed animal but fails to show the evidences of estrogenic hormone secretion. We have also reported that the tumor-bearing calorie restricted mouse does not respond to gonadotropin. The restriction involves only calories, the absolute amounts of protein, minerals and vitamins being normal.

In this study the mice, ovariectomized at 19–25 days of age and restricted in calories to 66%, were injected (at 13–15 months of age) subcutaneously with 0.1 mgm. corticotropin (Armour, Lot No. 32-D. 1 mgm. = 0.71 ± 0.09 mgm. La-1-A) daily for 19 days. Since no change had occurred in the vaginal smear they were then given 0.1 mgm. twice daily for 9 days. The animals remained consistently in anestrus before, during and after the injections. Smears were made daily for three weeks after the last injection. Approximately one month later the animals were sacrificed and found to have adrenal tumors but immature uteri and vaginae. It is evident that in calorie restricted ovariectomized C3H mice the tumorous adrenal, which is presumably the source of estrogen in the full-fed animal, is not responsive to corticotropin in the amounts used.

Visceral capillary permeability changes produced in the dog by anaphylactic shock and venous stasis. JOHN S. KIRK (introduced by K. E. Jochim). *Dept. of Physiology, Univ. of Kansas, Lawrence.* It has been shown by other investigators that an increased flow of thoracic duct lymph and liver engorgement similar to that observed in anaphylactic shock could be produced in the dog by clamping the hepatic veins. Comparison has been made between anaphylactic shock and conditions which cause hepatic engorgement with a fall in mean arterial blood pressure. These experiments were designed to make a further comparison of these conditions. Permeability alterations in the extrahepatic visceral capillaries drained by the portal vein (with the exception of the spleen), immediately following anaphylactic shock in the dog were compared with changes in the same area produced by the injection of histamine dihydrochloride and by mechanical occlusion of the hepatic veins and portal vein. Splenectomy was performed before each experiment. Determinations of hematocrit and plasma protein concentration (falling drop method) were made on arterial and portal venous blood. A loss of the larger molecules from the plasma was demonstrated in anaphylactic shock and following injection of histamine dihydrochloride within the first few minutes after the fall in mean arterial blood pressure. No loss of larger molecules from the plasma was demonstrable during the first few minutes after hepatic or portal venous stasis. From the hematocrits, the theoretical increase in plasma protein concentration in the portal venous blood resulting from fluid loss alone was calculated and compared with actual changes, in order to indicate whether or not there was any loss of protein from the vascular bed.

Differences in exocardial and endocardial electrograms of rabbit, dog and calf. BRUNO KISCH, FRANZ M. GROEDEL (by invitation) and PAUL R. BORCHARDT (by invitation). *From the Biological Laboratory, Fordham University, New York.* Electrograms were taken under Nembutal nacrosis simultaneously from the surface and the inside of the calf heart a few days after birth. These are compared with electrograms recently obtained with identical technique from rabbits and dogs by the same authors. They show similarities and some conspicuous dissimilarities. The ventricular complex obtained from the cavity of the left ventricle of the calf begins with a tall upstroke (R), that from the surface at the apex of the left ventricle with a deep downstroke, forming a QS. In the rabbit and dog it is exactly the opposite. The ventricular electrogram taken from the surface of the right auricle, from inside the carotid artery, aorta, right auricle and superior vena cava, in dog and rabbit, contains no or a small R, and a deep negative deflection (QR). In the calf, the corresponding electrograms and that obtained from inside the left auricle show no Q, a high R, while S may be present or absent. As in dogs and rabbits, it is possible to produce auricular fibrillation by the local application of acetylcholine to the auricular surface. In the calf the interval between its application and the start of fibrillation is greater; the fibrillation lasts longer than in dogs. Typical fibrillary waves occasionally may be present in the tracing taken on the surface of the right auricle but absent in the tracing from inside the left auricle.

The electrogram of the fish heart. By BRUNO KISCH, New York, N. Y. *From the Marine Biological Laboratory, Woods Hole, Mass.* Electrograms were taken from the surface and from the inside of the heart of Bone Fish and of Selachiens with semiunipolar leads. The voltage of electrograms registered in this way has the same range as that of dogs and rabbits. The electrogram of the auricle as well as of the ventricle shows a QRS complex, a T and sometimes a following U-wave. The duration of the QRS is normally 0.04 to 0.06 seconds but the QT distance has a long duration (0.4–0.8 sec.). The statement of the literature that in the fish's heart a-v conduction as a rule is slower and less good than v-a conduction could not be corroborated. The spread of the excitation wave starts synchronously on the inside and outside of the ventricle. The apex of the ventricle is activated later than the base, but the activity of the base lasts normally longer than that of the apex.

The bulbus cordis of Selachiens can become pacemakers of the heart and its electric activity can be registered. The one ventricle of fish is anatomically incompletely subdivided into a left and right apartement. The electrograms taken in each of these compartements may differ from each other remarkably. Potassium Chloride increases the rate of the actual pacemaker and transforms potential pacemakers into actual ones. Acethylcholinechloride and Mecholyl applied to the auricle slowed down the heart rate, but did not start auricular fibrillation.

Variations in skin temperatures of the feet and hands and the onset of sleep. N. KLEITMAN, A. RAMSAROOP (by invitation), and T. ENGLMANN (by invitation). *Dept of Physiology, Univ. of Chicago, Ill.* The sudden steep rise in the temperature of the feet one or two hours before (sometimes immediately before or after) the onset of sleep has been interpreted as evidence of peripheral vasodilation due to a decrease in sympathetic activity, manifesting a "vegetative preparedness for sleep". Our observations of skin temperatures of the toes, soles, and legs, as well as the fingers, palms and forearms (or cheek), under a variety of conditions, while confirming the reported facts concerning the feet, do not support the interpretation given to them. In general, skin temperature fluctuations in the toes are greatest, in the soles, intermediate, and in the calves, least. The same differential applies to the corresponding skin areas in the upper extremities, the absolute changes, however, being smaller. Following a meal, if there is a rise in oral temperature, there is a concomitant increase in toe and finger skin temperatures. Muscular relaxation, when it leads to a lowering of the body temperature, is usually accompanied by a drop in finger temperature and a simultaneous rise in toe temperature. In the pre-sleep hours of the evening there is thus a combination of two toe skin-temperature raising influences: meal and muscular relaxation. It therefore seems that the feet, especially the toes, exhibit vasomotor changes that are not characteristic of the skin as a whole, not even of the skin of the extremities, and changes in foot temperatures cannot be looked upon as variations in general sympathetic activity.

On naturally occurring porphyrins in the root nodules of leguminous plants. HEINRICH KLÜVER. *The Division of the Biological Sciences, University of Chicago.* The fluorescence spectrum of the root nodules of various leguminous plants containing red or brown pigments exhibits an emission band at about 620 mμ. The porphyrin which we have extracted from the nodules of the soybean has the characteristics of a coproporphyrin. The spectrochemical evidence has been derived from data on solubility, HCl number, and measurements of the fluorescence spectra of the free porphyrin and its methyl ester in various solvents. Previous work (Kubo, Keilin, Virtanen) has established the occurrence of hemoglobin in the root nodules of every leguminous plant so far examined. It becomes possible, therefore, to study a porphyrin-hemoglobin system in the absence of numerous factors influencing or complicating the relations between porphyrins and hemoglobin in the animal organism. Our preliminary studies of root nodules of the red kidney bean have revealed the presence of large amounts of a porphyrin which spectroscopically is practically identical with coproporphyrin, but which in other respects behaves like the Waldenström type of uroporphyrin. Future research will be necessary to determine whether the legporphyrins include a hitherto undescribed porphyrin. The physiological rôle of the porphyrins occurring in various organisms is not exactly known. The question must be raised whether the porphyrins present in the root nodules are directly or indirectly involved in the process of symbiotic nitrogen fixation, particularly in the earlier stages. The use of isotope methods for future investigations of this problem is suggested.

A comparison of the effect of testosterone propionate and "growth hormone" on the body and organ weights and composition and the arginase and phosphatases of the kidney and liver of the castrated male mouse. CHARLES D. KOCHAKIAN and CONSTANCE E. STETTNER (by invitation). *Department of Physiology and Vital Economics, University of Rochester, Rochester 7, New York.* White male mice were castrated at 17–19 grams body weight and approximately one month later, separate groups were treated for 10, 20 and 34 days as follows: a) injected daily with 1 rat growth unit of anterior pituitary growth hormone (Parke, Davis & Co.) b) implanted subcutaneously with a 14 to 15 mg. pellet of testosterone propionate and c) both treatments simultaneously. The mice treated with the growth hormone preparation showed approximately the same increase in body weight as those treated with the androgen pellet. When the two hormones were administered simultaneously there was a summation effect on the increase in body weight. The growth hormone increased the kidney and liver in proportion to the effect on body weight. The androgen produced the well known effects on the organs. The kidney weight was increased about three times that produced by the growth hormone. There was no summation of these responses when the two hormones were administered simultaneously. Both hormones increased the total amount of protein and water in the carcass and the organs and decreased the fat of the carcass. The increased amount of protein

was similar for both hormones except that the androgen diverted a large portion, 15–20 percent, to the seminal vesicles and prostates. The growth hormone caused a slightly greater decrease in water content and a much greater decrease in carcass fat. The simultaneous administration of the two hormones caused a summation of these effects. The androgen produced the expected changes in the enzymes of the kidney. The growth hormone increased all of the enzymes in proportion to the increase in mass of the organ. When the two hormones were given simultaneously there was a remarkably smaller increase in the arginase activity of the kidney; the phosphatases were not affected. The concentrations of enzyme activities in the liver were not affected by any of the treatments.

Acute pulmonary edema produced by ammonium salts. HAROLD KOENIG and RUTH KOENIG (introduced by W. F. WINDLE). *Anatomical Laboratories, University of Pennsylvania School of Medicine.* During the course of investigation of another problem, in which guinea pigs were administered ammonium chloride daily by gavage to produce chronic acidosis, it was noted that animals occasionally succumbed to an acute pulmonary edema. The characteristic sequence of events transpires during a period of fifteen to sixty minutes following the administration of 0.06 to 0.12 gm. of ammonium chloride per 100 gm. of body weight. The subcutaneous, intramuscular, intraperitoneal, intravenous and enteric routes of administration are all effective. Following a period of dyspnea; coma, muscle fasiculations and generalized clonic and tonic convulsions supervene. A serous or serosanguineous fluid usually exudes from the nostrils before exitus. The lungs are typically swollen and present areas of gross subpleural hemorrhage and congestion. Copious edema fluid can be expressed from the cut survace of the lung. The right heart and entering veins are greatly distended. Other ammonium salts in equivalent amounts in respect to the ammonium ion will produce pulmonary edema. Rats and cats also are susceptible. Bilateral cervical vagotomy, atropinization, and barbiturate anesthesia do not prevent the pulmonary edema although they occasionally alter the typical pattern of events. Analysis of edema fluid from one cat revealed it to be a transudate with no protein, indicating that an increase in pulmonary capillary permeability is not the prime factor producing this edema. However, the fluid is blood-tinged in many cases and probably contains serum proteins that leak out of the capillaries after anoxia sets in.

Secretion of mucin in response to sham feeding and histamine stimulation. S. A. KOMAROV, HARRY SHAY, HERMAN SIPLET (by invitation). *Fels Research Institute, Temple University, School of Medicine.* Although it has been long recognized that mucin in solution is one of the principal constituents of acid gastric juice, (Babkin, B. P., Am. J. Surg., **7**, 498 (1929); Canad. Med. Assn. J. 28, 134 (1931); Webster, D. R., and Komarov, S. A.; J. Biol. Chem., 96, 133 (1932); Komarov, S. A., J. Biol. Chem., 109, 177 (1935)) the mechanism of its secretion is little understood largely due to the lack of reliable quantitative methods for determination of this substance. With the aid of a newly developed method for mucin estimation, (S. A. Komarov, H. Siplet, and H. Shay; Federation Proceedings, Vol. 6, No. 1, March, 1947) it has been possible to compare the secretion of mucin in addition to pepsin, acid and protein nitrogen under various conditions of gastric stimulation. Experiments performed on three dogs with gastric fistulae and oesophagotomy demonstrated conclusively that sham feeding stimulates the secretion of mucin as effectively as it does that of acid and pepsin. Furthermore, an analysis of the curves for concentration and output of the major constituents shows each of them to behave independently, an indication that the respective sets of secretory cells respond in a selective manner to the same stimulus transmitted through the vagus nerves. Experiments with histamine showed that the parietal cells alone are stimulated by this substance. The output of mucin remained constant at the level of resting stomach secretion, while pepsin secretion in many experiments was actually inhibited.

Dermatomal autonomic activity in relation to segmental motor reflex threshold. IRVIN M. KORR and MARTIN J. GOLDSTEIN (by invitation). *Still Memorial Research Trust, Kirksville, Missouri.* Enduring differences in segmental reflex thresholds involving the spinal extensor motoneurons have been demonstrated in man (Denslow, J. Neurophysiol. *7:* 207, 1944). The low threshold segments appear to be those in which a relatively large portion of the motoneurons are maintained in a state of facilitation due to chronic bombardment by impulses from segmentally related structures (Denslow, Korr and Krems, Amer. J. Physiol. **105:** 229, 1947). In the present investigation evidence has been obtained that the facilitation extends to the cells of the intermediolateral column in the corresponding segments since measurements of electrical skin resistance indicate segmental differences in sweat gland activity which are related to the motor reflex thresholds. Electrical conductivity of the skin of the back was measured in our subjects by a convenient modification of the dermohmmeter. Under the conditions of our experiments most of the skin of the back has a resistance of 5,000,000 ohms or more. However, portions of dermatomes, and oc-

casionally entire dermatomes, related to segments with reduced thresholds have markedly reduced electrical resistance, often as low as 20,000 ohms. The largest, most constant and most reproducible differences in skin conductivity, related to segmental motor reflex thresholds, are found in the midline, over or near the vertebrae. Areas with reduced resistance are often hyperesthetic and some may have the characteristics of trigger areas. It is concluded that in segments with chronically reduced motor reflex thresholds, at least some of the preganglionic sympathetic neurons of the same segments are also maintained in a state of facilitation.

Influence of unilateral adrenalectomy and sympathectomy on homolateral renal function. J. P. KRISS, P. H. FUTCHER and M. L. GOLDMAN (introduced by H. A. SCHROEDER). *Department of Internal Medicine, Washington University School of Medicine, Saint Louis, Missouri.* In 1914 Cow reported that unilateral adrenalectomy caused increased excretion of urine by the homolateral kidney; he also described direct vascular connections between adrenal and kidney. In 1919 Marshall and Kolls reported that changes in renal function following unilateral adrenalectomy were occasioned by interruption of autonomic nerves during adrenalectomy.

The excretion of water, chloride, mannitol, and sodium para-aminohippurate was studied in dogs under nembutal anesthesia with both ureters catheterized. The excretion of all these substances by the left kidney exceeded that by the right kidney in 3 dogs several weeks after left adrenalectomy and in 1 dog immediately after left adrenalectomy. Increased excretion of these substances by the left kidney was also noted in 2 dogs with intact adrenals several weeks after left supradiaphragmatic sympathectomy and in 1 dog with intact adrenals immediately after the same operation. No similar difference in function of the 2 kidneys was observed in 2 dogs several weeks after bilateral supradiaphragmatic sympathectomy and left adrenalectomy.

Our results support the conclusion that unilateral sympathectomy and unilateral adrenalectomy produce the same effect upon those aspects of renal function which we investigated. In most, but not all, instances, the increase in glomerular filtration rate (mannitol clearance) sufficed as a possible explanation for the increased excretion of water and chloride by the kidney homolateral to the site of the operation.

The adrenalin sensitivity of the denervated dog kidney. W. G. KUBICEK, R. B. HARVEY and F. J. KOTTKE. *University of Minnesota, Minneapolis.* Renal plasma flow (RPF), glomerular filtration (GF) and filtration fraction (FF) were determined by means of para-aminohippurate and creatinine clearance respectively. After control values were obtained epinephrine hydrochloride (Winthrop) was administered by a continuous intravenous infusion at a rate sufficient to elevate the diastolic blood pressure approximately 25 mm. Hg. In most instances 4 to 6 mgm. of adrenaline per hour produced the desired results. Nausea and vomiting occurred in some cases if the adrenalin infusion rate became excessive. In dogs with normal renal nerves no significant change was noted in RPF, GF or FF. Dogs with denervated kidneys showed a marked reduction (40 to 50 per cent) in RPF and GF with no significant change in FF. The data indicate that the dog with normal renal innervation can maintain a constant renal function during a large increase in arterial blood pressure produced by the above procedure while the increased sensitivity to adrenalin in the denervated kidney produces a vasoconstriction of greater proportion than the increase in blood pressure. This factor may be of importance in connection with sympathectomy in man.

Effect of sinus gland extract on the rate of phosphorylation of arginine. ELOISE KUNTZ (introduced by IRWIN SIZER). *Arnold Biological Laboratory, Brown University.* Many types of investigation have demonstrated the endocrine nature of the crustacean sinus gland. An investigation of the effect of crayfish sinus gland extract on the rate of transfer of phosphate from adenosine triphosphate to phosphoarginine by muscle homogenate was made. Homogenates prepared from the abdominal muscles of crayfish were pre-incubated with an aqueous extract of sinus gland, then incubated with a substrate containing arginine and adenosine triphosphate in carbonate buffer at pH9. Magnesium chloride and potassium chloride were also added. The control experiments contained all components except sinus gland extract. An acceleration of phosphate transfer from adenosine triphosphate to arginine of the order of two to four times occurred in the presence of sinus gland extract.

Effect of lactogenic hormone on estrogen response of immature rats. ANNETTE LABELLE (by invitation) and RICHARD TISLOW. *Biological Research Laboratories, Schering Corporation, Bloomfield, New Jersey.* When lactogenic hormone (prolactin) was administered to immature rats in which corpora lutea had been induced by the administration of human chorionic gonadotrophin, no vaginal cornification was produced by an otherwise effective dose of estradiol benzoate.

A study of the ankle jerk in myxedema. EDWARD H. LAMBERT, LUIS O. MEDEROS (by invitation) and MAVIS P. KELSEY (by invitation). *Sections on Physiology and Medicine, Mayo Clinic and Mayo Foundation, Rochester, Minnesota.* The strength and duration of the tap on the tendon, muscle action potential and muscular tension have been re-

corded simultaneously in studying the ankle jerk of 20 normal subjects and 25 patients with myxedema. The duration of the contraction and relaxation phases of the muscle response were 0.14 (0.11–0.19) and 0.33 (0.19–0.49) seconds, respectively, in normal subjects (BMR +4 to −12), but were 0.23 (0.19–0.28) and 0.74 (0.43–1.09) seconds, respectively, in myxedema (BMR −16 to −39). The duration of contraction was generally inversely proportional to the BMR. It decreased as BMR increased in hypothyroid patients given desiccated thyroid. It increased as BMR decreased in patients developing myxedema following administration of radioactive iodine. The total latent period (0.035 seconds) and the contour and duration of the muscle action potential were not significantly different in myxedema and normal subjects. However, the duration of a single muscle twitch following direct electrical stimulation was greater in myxedema than in normal subjects. The temperature of the muscle in myxedema was 0.5–1.0°C. lower than that in normal subjects, but it was necessary to reduce the temperature of normal muscle approximately 10°C. to slow its response to the degree found in myxedema. Furthermore, the ankle jerk in myxedema differed from that of normal muscle cooled to 25–28°C. in that (1) the action potential was not prolonged as it was in cooled normal muscle, and (2) a "shoulder" was prominent in the relaxation phase of the response of myxedema muscle, but not in cooled muscle.

Studies on the nutritional effects of heated fats. A. LANE (by invitation) and A. C. IVY. *Dept. of Clinical Science, Univ. of Illinois College of Medicine, Chicago.* The work of several investigators suggests that fats heated at high temperatures are nutritionally harmful and may be carcinogenic. The following experiment was designed to study the effect of heated fat as compared to unheated fat on the nutrition of rats. Two diets composed of a homogeneous mixture of bread, milk and lard which in one diet was heated to 350°C. for thirty minutes and unheated in the other diet were fed to young adult white rats. Four synthetic diets, two adequate for rat nutrition, the fat content of one being heated (350°C. for 30 min.) and two diets high in fat (43%) and low in protein (8%) one of which contained heated fat were fed to four groups of rats. Weekly body weight and daily food intake were recorded, and at intervals animals from each group were killed, tissues taken for histological study and total fat determinations run on livers. There is little difference in food intake between rats on unheated and heated fat with the bread, milk and lard diets; both groups show skin lesions and eye signs suggestive of vitamin A deficiency. On the synthetic diet, the group eating the "adequate heated fat diet" ate slightly less and body weight curve is slightly below the "unheated fat group". In the group eating high fat which has been heated, the food intake and body weight curve are below the group eating the same diet with unheated fat content. These studies are being continued.

Cyclic changes in the respiratory center. M. G. LARRABEE and ROBERT HODES. *Johnson Foundation, University of Pennsylvania.* Progressive changes in the respiratory centers, occurring during the respiratory cycle, were tested by afferent impulses over the superior laryngeal nerve. In several experiments the minimum numbers of afferent volleys required to stop inspiration were determined at various times during inspiration. Thus measured, the threshold for stopping inspiration fell linearly from early in inspiration to reach zero at the end of inspiration. Thus changes which normally terminate inspiration develop progressively throughout inspiration, and follow a simple time course. In other experiments constant numbers of afferent volleys were initiated at various times during expiration; the delay caused in the next inspiration was observed. The maximum delay resulted from afferent volleys initiated just before the normal start of inspiration. Delays due to afferent volleys earlier in expiration were expressed as percent of the maximum delay, in animals breathing at widely different rates. Delays due to afferent volleys one second before the end of expiration were smaller fractions of the maximum, the shorter the animal's natural expiratory pause. Delays due to afferent volleys at the start of expiration were always between 39 and 47% of the maximum, with no consistent change with duration of expiration. This constant reduction in inhibitory effect, during a time equal to one expiration, may be explained by assuming equal rates of decay for a) the central effect produced by afferent impulses and b) for inhibitory effects which normally develop during inspiration and subside during expiration. This further suggests some mechanism in common for the two effects.

Estimation of the rate of antidiuretic hormone secretion in normal man. HENRY D. LAUSON, HOWARD A. EDER (by invitation), FRANCIS P. CHINARD (by invitation), GEORGE C. COTZIAS (by invitation) and ROGER L. GREIF (by invitation). *Hospital of the Rockefeller Institute for Medical Research, New York, N. Y.* From studies on dogs with experimental diabetes insipidus, Shannon (J. Exp. Med. **76:** 387, 1942) concluded that the normal rate of secretion of antidiuretic hormone in intact 10 to 15 kg. dogs is equivalent to the intravenous administration of from 1 to 5 milliunits of pituitrin per hour. In order to make the same kind of estimate in a normal 70 kg. man, the endogenous secretion of antidiuretic hormone was inhibited by rapidly drinking 1500 ml. of water and the inhibition maintained for 8 hours by drink-

ing water in volumes equal to the urine output. After a 2 hour control period of steady maximal diuresis, commercial Pitressin was infused intravenously at the constant rates of 7.5, 16.4 and 50 milliunits per hour during 3 consecutive 2 hour periods. On another day under the same regimen, 333 milliunits per hour were administered for 2½ hours. The endogenous creatinine U/P ratio (urine concentration/plasma concentration) was taken as the index of renal tubular water reabsorption. Assuming that after 1½ hours of constant infusion, equilibrium has been attained between the rate of Pitressin inflow and the rate of its removal from the blood, the U/P ratio during the next half hour measures the renal response to the given infusion rate. For the rates of 0, 7.5, 16.4, 50 and 333 milliunits per hour, the following U/P ratios were found at equilibrium: 7.2, 80.8, 138, 183 and 134, respectively. Per kg. body weight, these results are essentially the same as obtained by Shannon in dogs with diabetes insipidus.

High protein diets and testosterone propionate as related to plasma and liver proteins in rats. JAMES H. LEATHEM. *Bureau of Biological Research, Rutgers University, New Brunswick, New Jersey.* The influence of a high protein diet on adrenal size and function has been studied in adult and young rats over a 20–25 day period. Diets of 78% casein or lactalbumin were the high protein diets used whereas diets of 18% protein or fox chow were considered as control diets. Body weight increase was retarded by the high protein diets, an effect which was more pronounced in adult rats by the use of lactalbumin. A modest hypertrophy of the adrenals was noted and a significant increase in kidney and liver weight was obtained. If the plasma protein concentrations are used as a criterion of adrenal function then the modest adrenal hypertrophy noted did not alter function. This is indicated by the fact that total plasma protein, albumin and globulin concentrations were not influenced by diet although NPN was increased by a high protein diet. The increase in liver size, however, was accompanied by an increase in total liver protein in the body and an increase in protein percentage of the dry weight. Water content of the liver was not changed. Since testosterone propionate is known to cause urinary N_2 retention, the effect of this androgen in combination with varied diets was studied. Testosterone propionate (Perandren, Ciba) was administered three times weekly in dosages of 1.0 mg. This amount of androgen did not influence organ weight, plasma protein concentrations or liver protein content.

The effect of intravenous sodium cyanide on the electrocardiogram. ALFRED LEIMDORFER. *Department of Psychiatry, University of Illinois.* Intravenous injection of NaCN (0.2–0.8 mg./kg.) causes prompt, brief, reflex slowing of the heart in cat, monkey and man. In the cat this depends on the carotid bodies and vagi. During the first slowing, high take-off waves, similar to those seen in the acute stages of coronary occlusion, appeared in the cat alone. A second slowing, with increased T-P and P-R intervals, follows. With toxic doses in the cat (0.9–2.4 mg./kg.) this passes into extreme bradycardia and arrhythmia. There also appeared in cat, monkey and man increased amplitude of T waves and elevation of S-T segment. These and other typical anoxic changes varied with the dose of NaCN. With the largest doses in cats signs of myocardial failure appeared in the form of severe arrhythmias, a-v and intraventricular blocks, extreme low voltage, ventricular flutter and fibrillation.

Electroencephalographic analysis of action of amidone, morphine and strychnine on the central nervous system. ALFRED LEIMDORFER. *Department of Pharmacology, University of Illinois, College of Medicine, Chicago.* In cats, amidone-HCl induces changes in the electroencephalogram (EEG) (of the parietal and occipital lobes): with 3 mgm./kgm. of body weight i.v., there appear transitory diphasic spikes alternating with a decrease in frequency. After the i.v. injections of larger doses (18 mgm. per kgm. over a period of 6½ hours), transitory, high voltage, high frequency-discharges, indicative of seizures, occur. Following the initial change, the control activity returns. After the i.v. injection of still larger doses (22.5 mgm. per kgm. within 6½ hrs.), the electrical activity of the brain is abolished. No changes in the electrospinogram are obtained. In cats, morphine sulfate (5 mgm./kgm., subcut.), produces a slight transient increase in the frequency of the EEG. The i.v. injection of large doses (40 mgm. per kgm.), induces a great decrease in the frequency and voltage of the EEG, after the additional i.v. injection of 40 mgm. per kgm., about an hour later, the brain waves disappear. The injection of morphine rarely produces the appearance of diphasic spikes in the EEG and occasionally the occurrences of tetanic waves in the electrospinogram. In cats, strychnine sulfate (0.5 mgm./kgm. subcut. or i.v.) produces a burst of tetanus-waves in the electrospinogram. The i.v. injection of large doses (2.5 mgm./kgm.) evokes characteristic diphasic spikes in the EEG. Section of the spinal cord shows that tetanic waves arising from different parts of the spinal cord and the spikes in the EEG are independent in origin.

Auditory and visual areas of the cerebral cortex of the rat. D. H. LEMESSURIER (introduced by C. N. WOOLSEY). *Dept. of Physiology, Johns Hopkins University, School of Medicine, Baltimore 5, Md.* The evoked potential method has been used to define the auditory and the visual areas of the rat's cerebral cortex. The potential changes produced

in the cortex either by photic stimulation of the eye or by click stimulation of the ear are very large and may range from 1500 to 2500 microvolts under prolonged pentobarbital anesthesia. The main auditory area, centered over Krieg's area 41, is approximately 4 mm. wide dorsoventrally and about 5 mm. long antero-posteriorly. There is also another area which can be activated by sound. This apparently corresponds to Tunturi's third auditory area. It is closely related to the face subdivision of somatic area II (see Woolsey, and LeMessurier, this issue). The visual area corresponds approximately to Krieg's areas 17, 18 and 18a. Both eyes project to this area, but the responses evoked by stimulation of the ipsilateral eye are large only in the central part of the field activated by the contralateral eye. There is a slight degree of overlap between the visual and auditory areas posteriorly. Both areas overlap somewhat the adjacent portions of the somatic areas. Whether these overlaps mean anatomical intermingling or result from the method has not been decided. In any case there is no cortex between the various receiving areas which could constitute any appreciable associational cortex. The nature of the overlap encountered does not suggest that associative functions are subserved by the overlap.

The secondary blood pressure rise and tachycardia occurring after the injection of epinephrine. R. LENEL (introduced by S. RODBARD). *From the Cardiovascular Department, Research Institute, Michael Reese Hospital, Chicago, Illinois.* We have observed that during the course of the primary pressor action of large doses of intravenous epinephrine, injected in trained unanaesthetized dogs, a secondary rise in blood pressure of the order of 75/75 mm. Hg above the primary peak may occur. The entire secondary rise in pressure occurs within a beat or two, and it is followed immediately by a paroxysm of ventricular tachycardia which may persist for a few seconds or for as much as two minutes. The incidence of the occurrence and the duration of this effect seen with various doses of epinephrine is given in the table.

Epinephrine injected	Animals with secondary rise	Average rise	Duration of tachycardia
mg.	%	*mm. Hg*	*sec.*
0.01	0	0	0
0.1	22	63	32
0.5	50	66	32
1.0	75	96	118
2.0	80	78	135

The effect is inhibited by atropine, dibenamine or by pentobarbital anaesthesia. It is partially inhibited by morphine.

Chemical and physiological changes during water tolerance tests in patients with cirrhosis. STEPHEN LESLIE (by invitation), GEORGE H. STUECK, JR. (by invitation), HAROLD M. SHORR (by invitation) and ELAINE P. RALLI. *Department of Medicine, New York University College of Medicine.* Water tolerance tests were done in patients with cirrhosis of the liver, both with and without ascites, and in normal subjects. The tests were done by administering 1500 ml. of tapwater orally. The plasma volume was determined prior to the test. Blood and urine samples were taken at intervals during the tests. The hematacrit, total protein, albumin, globulin, sodium and chloride were determined on the serum. The rate of urine flow was accurately determined by collecting the urine by catheter and the specific gravity and the chloride content were done on the urine samples. To rule out the effect of intra-abdominal pressure on the rate of urine flow some tests were done on the same patient both before and after a paracentesis. In other patients repeated tests were done during periods when ascites was present and when ascitic fluid had ceased to reaccumulate. The results showed that in patients with cirrhosis of the liver there is a marked retardation of the rate of urine flow which occurred even when there was no evidence of ascites. The plasma volume as determined by the dye method was increased in all of the patients. During the test the chloride content of the serum fell regularly. The other fractions determined showed certain variations which will be presented.

The path of suppression in the spinal grey matter. JEROME Y. LETTVIN (introduced by W. S. MCCULLOCH). *Department of Psychiatry, University of Illinois.* In the cat's spinal cord, soaked in nembutal in situ to prevent transynaptic activity, impulses evoked by electrical stimulation of the bulbar reticular suppressor mechanism were recorded on one occasion from the medial internuncial pool of the ventral horn, and regularly from the lateral portion of the intermediate pool of internunnials. Even without soaking in nembutal, they have never been found in any other position in the spinal grey. Stimulation in this lateral portion of the intermediate internuncial pool with 30 micro-amperes completely inhibited the knee jerk. Pick up and stimulation of cord were through concentric bipolar microelectrodes separated less than half a millimeter. Their positions were histologically determined by Dr. R. S. Snider.

The disposal of intravenously administered amino acids by normal, hepatectomized, depancreatized and hyperthyroid dogs. R. LEVINE and N. G. SCHNEEBERG (by invitation). *Department of Metabolism and Endocrinology, Research Institute, Michael Reese Hospital, Chicago, Illinois.* Dogs under morphine sedation, were injected intravenously with varying amounts of amino acids at a constant rate for 2 hours. Amino acid and urea

levels were followed in the blood and urine for 6–8 hours, beginning with a control period of one hour. During the experimental period the total extra nitrogen excretion amounted to 8–12% of the amount administered. 88–92% of the amino acids injected were retained. Neither the liver nor the pancreas seem to be necessary during this phase of amino acid assimilation. Pretreatment with thyroxine did not increase the N excretion during this period over the values in the normal animal.

Quantitative examination of sensibility in peripheral nerve injuries. F. H. LEWEY. *The Peripheral Nerve Study Center, Hosp. of the Univ. of Pennsylvania.* Threshold examinations for returning sensitivity following nerve injuries by means of v. Frey's hairs and Head's algesimeter are satisfactory as long as improvement is progressing. However, the results become equivocal when no improvement of threshold values is apparent three months after the last examination. The conclusion might be drawn that the nerve had met an obstacle and should be explored surgically. Yet, threshold determination is only one parameter of functional return of sensitivity. The number of touch and pain fibers which at a given time have made contact with their peripheral end organs is another. Careful sensory examination requires 1) counting of the number of touch and/or pain points per cm^2, present in a previously anesthetic and analgetic area, and 2) their threshold in terms of grams of pressure. To give a practical example: In the analgetic area of an injured radial nerve on the back of the hand one pain point p/cm^2 was discovered on schedule after nerve suture. These points felt pin prick under 8 g of pressure. Three months later, the threshold was still 8 g, and the patient seemed to be doomed for another operation. Fortunately, count of the pain points showed that their number had increased from 1/cm^2 to 20/cm^2 indicating that many more nerve fibers had arrived in the skin and made contact with their end organs, although their maturition had lagged behind. The operation was called off and the continued progress of recovery justified the decision.

Nature of increased serum β-globulin content in malignant hypertension and diabetic retinopathy. LENA A. LEWIS and IRVINE H. PAGE. *Research Division Division of the Cleveland Clinic Foundation, Cleveland, Ohio.* A consistent, absolute and relative increase of plasma β-globulin concentration has been found by electrophoresis of samples from patients suffering from malignant hypertension (Lewis and Page) and from diabetic retinopathy (Lewis, McCullagh and Schneider). Since this abnormality of electrophoretic pattern might arise from an increase in lipid loosely bound to β-globulin and migrating therewith, post-absorptive samples of serum from such patients were re-examined after cold acetone and ether extraction (Blix). Electrophoresis was done using phosphate buffer pH 7.8, ionic strength 0.16μ. The total protein concentrations were above the lower limits of normal.

As was expected, extraction decreased the area of the β-globulin peak. However, the relative concentration of β-globulin after extraction was consistently greater in sera from cases of malignant hypertension and diabetic retinopathy than in normal sera after or even before extraction. In lipid-extracted sera from normal subjects, the β-globulin area represents 12.0 to 15.4 (mean 13.6) per cent of the total protein shadow, while in patients suffering from malignant hypertension the range of β-globulin after extraction ranged from 16.1 to 32.8 (mean 20.2) per cent of total. These forms of cardiovascular disease are therefore shown to be associated with an increase in serum β-globulin content which is not due to loosely bound lipid. Although increased thymol turbidity has been related to high concentrations of β-globulin in liver disease, thymol turbidity was not increased in the form of hyper-β-globulinemia under study.

Adenosinetriphosphatase (ATP-ase) in nerve. B. LIBET. *Dept. of Physiology, The University of Chicago.* It is conceivable that a basic feature of nerve impulse conduction, the permeability change in the membrane, depends upon a structural change in certain membrane proteins. If the latter resemble the myosin system of muscle, they should be associated with ATP-ase activity. To test this hypothesis, measurements were made of the ATP-ase activity of the axoplasm, extruded from cleaned giant axons of the squid *Loligo pealii*, and of the remaining sheath portion. With the tissue homogenized in 0.58 M KCl, and the activity measured in 0.58 M KCl, 0.003 M $CaCl_2$, veronal buffer (pH = 7.4) and 0.003 M ATP, axoplasm splits phosphate from ATP at the rate of about 0.2 micrograms P/mg. wet weight in 30 minutes at about 26.5°C., while the average figure for the sheath is 19, about 100 times as much! Not only is the ATP-ase activity of the nerve fiber almost exclusively confined to the sheath portion, but the latter has an activity even greater than that of squid muscle. Contrary to the case of cholinesterase (D. Nachmansohn & B. Meyerhof, J. Neurophysiol., 1941, 5: 348–361), ATP-ase is not more concentrated in the optic ganglion than in the axon sheath. Nerve ATP-ase also resembles vertebrate muscle ATP-ase in its substrate specificity and in its sensitivity to Ca concentration, though it is considerably less sensitive to sulfhydryl poisons. More purified preparations of ATP-ase from nerve tissue will permit a fuller comparison of the physical and chemical properties of this enzyme with the myosin-ATP-ase system.

Effect of stretch on membrane potential in frog muscle. G. LING, (introduced by R. W. GERARD). *Dept. of Physiology, The University of Chicago.* The previously described technique (Graham and

Gerard, J.C.C.P. **28:** 99, 1946) for measuring the membrane potential of a single muscle fiber by inserting a micro-electrode has been improved with a capillary electrode of smaller diameter (app. 1 μ i.d., 3 μ o.d., measured 5 μ away from the tip) and gentle taper, the single fiber resting potential of frog sartorius is found to be higher and more constant. For nearly 200 fibers from 20 muscles, the average is 77.5 $\pm$ 4.7 mv., maximum, 90 mv., minimum, 60 mv. Stretching of striated muscle has been reported to effect heat production, respiration, pH, excitability, action and demarcation potential, etc. It was anticipated that stretch would alter the membrane potential. Measurements on many fibres from some twenty muscles, however, showed no significant differences between values obtained obtained before, during or after stretching muscles to length between 115 and 170% of the unstretched value. This is true even though muscles stretched to 170% remain some 10% longer than normal when released.

Effects of protein and fluid consumption upon plasma volume and circulating protein in the rat. Richard W. Lippman, (introduced by T. Addis). *Department of Medicine, Stanford University School of Medicine.* Rats were fed upon diets which varied from 3% to 77% in protein content, and contained variable quantities of fluid and salt. At the end of the experimental periods, 17 hours and 7 days, blood and plasma volumes were determined by a dye method, and total serum protein was determined by the biuret method. Total circulating protein was calculated from the measured data. Results were compared with normal for rats of the same size, as previously reported, in order to avoid errors inherent in surface area calculations. Changes in blood and plasma volumes were in no instance striking. Fluid diet with added salt produced a transient 10% increase in plasma volume. Blood volumes were increased only in groups which lost weight during the experiment, and then in such proportion as would be expected from the relative stability of the total erythrocyte mass over short periods. The high protein diet produced only slight increases in blood and plasma volumes, and only 6% increase in total circulating protein, at the end of 7 days. All protein-deficient diets reduced total circulating protein 15–20% at the end of 7 days. The apparent contradiction between our data and that of Metcoff, Favour and Stare (J. Clin. Invest. 1945, 24, 82) is due to their mode of expression in terms of "unit" function per 100 sq. cm., as the lines relating blood and plasma volumes, and total circulating protein, to surface area in the normal rat do not intercept either axis at 0.

Two fractions of specific cholinesterase present in normal mouse brain. J. Maxwell Little. *Department of Physiology and Pharmacology, Bowman Gray School of Medicine of Wake Forest College, Winston-Salem, North Carolina.* Mouse brain homogenate [NaHCO, buffer, pH 8.5] was centrifuged. The supernatant had 15–20 per cent and the resuspended precipitate had 80–85 per cent of the cholinesterase activity of the uncentrifuged homogenate. The supernatant from a distilled water homogenate [pH 6.8] contained 90–95 per cent of the uncentrifuged activity. Adjustment of distilled water homogenate to pH 8.5 by NaOH diminished the supernatant activity due to enzyme destruction; varying the NaCl molarity up to 0.075M before centrifugation resulted in a shift of activity from supernatant to precipitate, beyond this there was no further change.

The activity pattern of the supernatant and precipitate fractions was that reported for specific cholinesterase with the following substrates [0.015M]: acetylcholine, acetyl-β-methylcholine, propionylcholine, benzoylcholine and ethyl butyrate. There was no significant difference in the activities of the two fractions with acetylcholine when tested at pH 6.5, 7.0, 7.5 and 8.0 [optimum pH 7.5]. Incubating the supernatant and precipitate fractions obtained by centrifuging a distilled water homogenate made 0.15M with NaCl resulted in the following respective changes expressed as per cent of the activity of the unincubated fractions: 60°, 15 minutes—0 per cent, 0 per cent; 53°, 15 minutes—86 per cent, 93 per cent, 30 minutes—80 per cent, 72 per cent, 45 minutes—78 per cent, 50 per cent, 60 minutes—76 per cent, 33 per cent. Mouse brain contains two fractions of specific cholinesterase, one of which is precipitated from a water homogenate at a NaCl molarity of 0.075 or greater. The two fractions differ in their heat labilities.

Action of antacids in the human stomach; Results with zirconium phosphate. A. Littman (by invitation) and M. I. Grossman. *Dept. of Clinical Science, Univ. of Illinois College of Medicine, Chicago.* To study the effect of antacids on the volume of the gastric contents as well as on acid concentration a new technique was devised. Following histamine injection and introduction of antacid into the stomach tube, aspirations were made by the method introduced by Bloomfield and Keefer for study of the gastric secretory response to the alcohol test meal. The gastric contents were aspirated completely at 10 minute intervals, the volume measured, and all but a 10 cc. sample promptly returned to the stomach. The usual pH and "free" and "total" acid determinations were made on the samples. The effects of a thixitropic aqueous suspension of zirconium phosphate and aluminum hydroxide gel were compared, using 100 cc. volumes of each. In patients having a large acid output following histamine in control experiments, aluminum gel was shown definitely to be superior.

In patients having low acid response differences were small. It was thus shown that evaluation of the efficiency of gastric antacids by such a method requires consideration of the levels of acid output in control experiments as related to the dose of antacid used in the tests.

"Cortical instability": a study of frequency effects. ROBERT B. LIVINGSTON (by invitation) and JOHN F. FULTON. *Laby. of Physiology, Yale University School of Medicine, New Haven, Conn.* T. Graham Brown and C. S. Sherrington (*Proc. Roy. Soc.* 1912, **B85:** 250–277) and A. S. F. Leyton and C. S. Sherrington (*Quart. J. Exper. Physiol.* 1917, **11:** 135–222) called attention to the "instability" of single cortical points in the motor area. Repeated stimulation of a single point sometimes gave inhibition, sometimes facilitation; reversals of response were also encountered, i.e. the same point might give flexion of a digit at one time and extension the next. E. P. Boynton and Marion Hines (*Amer. J. Physiol.* 1933, **106:** 175–182) stimulated a cat and two macaques and reported evidence that the "instability" of the motor cortex was related to the frequency and intensity of stimulus. P. Bailey and G. von Bonin (*Trans. Amer. Neurol. Assn.* 1946, 89–93) noted that no facial movements could be obtained from the monkey face-area when stimulated at very low frequencies and that at these frequencies only movements of the tongue and vocal cords were evoked. O. Wyss (*J. Neurophysiol.* 1947, **10:** 315–320) provoked different central responses by stimulation of the vagus nerve in the monkey at different frequencies. F. Nulsen (personal communication) observed that alteration of frequency of stimulation of the anterior cerebellum changes response of cerebrally induced movements from inhibition to facilitation as the frequency scale is ascended. We have observed that frequency of stimulus is an important factor in eliciting autonomic responses to stimulation of the frontal areas in monkeys and man. A systematic study of certain cortical areas has been undertaken to learn the frequency range for different responses in relation to the different functional modalities of the particular cortical area.

Potentials of dorsal roots and related phenomena. DAVID P. C. LLOYD and A. K. MCINTYRE (by invitation). *Laboratories of The Rockefeller Institute for Medical Research, New York.* Potentials have been recorded in stimulated and adjacent dorsal roots. *In an adjacent root,* there are five consecutive deflections, only the last of which has achieved recognition as the so-called dorsal root potential. The first three (D.R. I, II and III) are an inverse electrotonic recording of the triphasic intramedullary spike potential of the travelling afferent impulses (negative, positive, negative at the central root lead). D.R. IV is positive, and overlapped by D.R. V; when cleared from it by asphyxia D.R. IV relaxes exponentially to half value within 3 msec. D.R. V is the familiar dorsal root negativity of Barron and Matthews. *In a transmitting root,* the spike potential is diphasic, followed by a negative potential of complex origin. The components have differential susceptibility to asphyxia. They are: the after-potentials of the afferent fibers, a negative potential similar to the positive D.R. IV of adjacent roots, and a negative D.R. V. Since the central projections of afferent fibers are mutually parallel, an E.M.F. generated by them must record electrotonically with opposite sign in transmitting and adjacent roots. D.R. IV, therefore, is referable to afferent fibers, and must be an electrotonic extension of the potential of terminating collaterals. D.R. V, of identical sign in transmitting and adjacent roots, must arise beyond the afferent fibers. The negative cord potential has has two components, dissociable by asphyxia. The more resistant of these is temporally identical with D.R. IV. It may be accounted for by a flow of current from the patent afferent fibers to the active collaterals. Positive cord potential and D.R. V are temporally identical.

Glomerular filtration rate in the adrenalectomized, salt fed rat. W. D. LOTSPEICH (introduced by R. F. PITTS). *Department of Physiology, Syracure University College of Medicine, Syracuse, New York.* Following clearance determinations in the intact animal, the kidney can be removed and isolated biochemical systems studied by tissue metabolism techniques. The rat is a suitable animal for this type of study. As a preliminary to such studies it was necessary to compare the rate of glomerular filtration in normal and adrenal insufficient rats. Twelve ad libitum fed controls, twelve adrenalectomized and six pair fed controls were studied. The creatinine clearance was used to measure glomerular filtration rate. After allowing all animals a three day acclimatization period on a "calf meal" diet, twelve rats were adrenalectomized. During the next two weeks records of body weight and food consumption were kept daily on all animals, and the adrenalectomized were allowed 1% sodium chloride to drink. Creatinine clearances were done during the third week when the adrenal insufficient animals were fully recovered and had established equality of body weight with their controls. It was found that glomerular filtration rate in the salt maintained adrenal insufficient rats was within the same limits as that of the normals. (Ad lib. fed controls averaged .731 cc./100 gram/min.; adrenalectomized averaged .901 cc./100 gram/min.; and the pair fed controls averaged .840 cc./100 gram/min.)

Heat loss and blood flow in the feet. L. LOVE (by invitation) and H. C. BAZETT. *Department of Physiology, University of Pennsylvania. Philadelphia, Pa.* Daily measurements of the heat loss of the foot,

blood flow and skin temperature have been made on two subjects living in a controlled temperature room for periods up to two weeks. Two conditions (approximately 33°C. and 21°C.) were used during the summer and again during the winter. During any single exposure to one temperature the only gradual change was in the temperature distribution along the foot. There were, however, considerable differences between three different periods of exposures to heat at different times of the year. The evaporative heat loss decreased after exposure to cold and there were associated changes in skin temperature, non-evaporative heat loss and blood flow. It has been possible to estimate the amount of heat lost from the foot as a percentage of the heat loss of the entire body. The area of the foot amounts to about 5% of the body surface. From this area 3.5% of the heat was lost in the cold and 6.5% in the heat. It has been found that Newton's law of cooling does not apply to the foot. For an equal temperature difference between skin and air about twice as much heat was lost in the heat as in the cold. This can be attributed either to a roughening of the skin in the cold, or to the change in temperature distribution already mentioned, since convective heat loss is affected by the curvature of the surface.

The effect of previous carbohydrate deprivation on the carbohydrate metabolism of isolated muscle. KNUD LUNDBAEK (by invitation) and JAMES A. F. STEVENSON (by invitation). (Introduced by C. N. H. LONG). *Dept. of Physiological Chemistry and Laby. of Physiology, Yale Univ. School of Medicine, New Haven, Conn.* Following starvation or carbohydrate deprivation the administration of carbohydrate reveals an impaired sugar tolerance and a very small or no increase in the respiratory quotient. The previous carbohydrate intake alone, not the total caloric intake, is the important factor in producing this state. To investigate the capacity of the peripheral tissues to handle carbohydrate at this time the isolated diaphragm of the rat was used. For two to three weeks one group of adult female rats was given a carbohydrate-free diet (80% fat, 20% protein by calories), and a second group a diet composed of 70% carbohydrate, 10% fat and 20% protein. Both diets contained adequate supplementary nutrients. At the end of this period the glucose uptake and glycogen synthesis were measured in the isolated diaphragm by incubation for three hours in a modified Krebs-Henseleit solution containing 200 mg.% glucose. There was no significant difference in the amount of glycogen synthesized. However, the glucose uptake of the diaphragms from the carbohydrate-deprived rats was little more than half that of the diaphragms of the rats which had received a high carbohydrate diet.

Concerning the nature of menstrual poison. DAVID I. MACHT. *Division of Pharmacology, Sinai Hospital, Baltimore, Maryland.* It is twenty-five years since the author published the first experimental proofs concerning the presence of a toxin in the blood of menstruating women. By means of quantitative plant-physiological experiments "menotoxin" was demonstrated in the blood, sweat, milk, tears, saliva and other secretions of such individuals. At that date the author concluded that menotoxin is a steroid closely related to oxycholesterol. Since then numerous hypotheses have been advanced as to its nature. In the present investigation the author tested by his phytopharmacological methods all such substances. The following bodies have been claimed as related to menotoxin: 1. various vasoconstricting substances 2. choline 3. di-methyl-amine 4. thyroxin 5. arsenicals 6. histamine 7. ascorbic acid 8. "necrosin." All these have been either prepared or secured by the writer and subjected to phytophamacological tests. None of them were found to be phytotoxic to an extent to be regarded as "menotoxin." On the other hand, numerous experiments with female sex-hormones and a large number of other steroids strengthen the original view of the author that menotoxin is related to cholesterol and oxycholesterol, and, while a separate entity, is closely allied to the female sex hormones. Certain biophysical observations obtained with sterols irradiated with ultraviolet and roentgen rays corroborated this view.

Thromboplastic effects of sulfa and penicillin combinations. DAVID I. MACHT. *Division of Pharmacology, Laby. of Sinai Hospital, Baltimore, Md.* The author has already described the powerful thromboplastic properties of penicillin, and its principal constituents (Science, **71**: 302, 1930). Inasmuch as both sulfa drugs and penicillin are often administered together especially to surgical patients, it is desirable to ascertain the effect of such combinations on blood coagulation. Experiments were made on rabbits and cats. Injections of sulfadiazine and sulfathiazole were made intraperitoneally or intravenously. Penicillin was administered either by vein or intramuscularly. Blood was secured either from carotid artery or by cardiac puncture. Neither sulfadiazine nor sulfathiazole affect coagulation time. Penicillin injections of either crystalline or amorphous variety quickly *decrease* coagulation time. This occurs both when the penicillin is given after the sulfa drug and when both are injected simultaneously. Too much emphasis can not be made on the importance of possible thrombo-embolic phenomena in patients receiving prolonged penicillin therapy, and the instituting of anticoagulant therapy.

Changes in skin temperature and blood flow of hand following ingestion of certain amino acids. MARTIN B. MACHT and ELIZABETH L. PILLION (introduced by H. S. BELDING). *The Quartermaster Corps, Climatic Research Laboratory, Lawrence,*

Mass. Skin and rectal temperatures, oxygen consumption, and blood flow through the hands of four healthy men were studied before and after oral administration of various amino acids. At an environmental temperature of 24°C. ingestion of glycine, in amounts of 1, 2, 3, and 4 grams/10 lbs. body weight, resulted in average hand skin temperature increases of 3.5°C., 4.8°C., 5.4°C., and 7.7°C., respectively, accompanied by similar, though less marked, increases in toe temperature and significant increases in blood flow through the hand as measured by venous occlusion plethysmography. Average maximum flows were 2.5 to 7 times the control flow. Increases in blood flow and hand skin temperature became apparent about 80 minutes after ingestion, being most marked approximately 180 minutes after ingestion. No significant changes in rectal temperature, or skin temperatures elsewhere were observed. At environmental temperatures of 18°C. and 30°C., no significant changes in skin temperatures or peripheral blood flow occurred after glycine administration. At an environmental temperature of 24°C., ingestion of phenylalanine effected increases in hand and toe temperatures and blood flow through the hand similar to those observed with glycine; oral administration of histidine resulted in slight but significant increases in hand skin temperature and blood flow. Ingestion of glutamic acid, tyrosine, leucine, and methonine had no effect upon skin temperature or peripheral blood flow. Although five of the seven amino acids caused definite increases in oxygen consumption, no consistent relationships between total oxygen consumption and skin temperature or peripheral blood flow were demonstrated.

The effect of acetyl-beta-methyl-choline on blood flow through the hand at low temperatures. MARTIN B. MACHT, MORTIMER E. BADER and ELIZABETH L. PILLION (introduced by H. S. BELDING). *The Quartermaster Corps, Climatic Research Laboratory, Lawrence, Massachusetts.* Utilizing a 0.2% solution of acetyl-beta-methyl-choline as the conducting medium in a metal venous-occlusion plethysmograph, this drug was passed through the skin of the hand by iontophoresis. With the plethysmograph serving as the positive electrode and a saline saturated negative electrode placed on the subject's back, a current of 10 milliamperes, derived from a 45 volt B battery through a 10,000 ohm variable resistance, was used to effect ionization. A thermostatically regulated refrigerating coil and heating unit within the plethysmograph permitted accurate control of hand skin temperature at any desired level throughout the experiment. All studies were condicted in a constant temperature room.

At a room temperature of 15°C. and a plethysmograph temperature of 10°C., blood flow was drastically curtained in the hands (less than 0.8 cc./100 cc. limb volume/minute). Iontophoresis of ABMC under these conditions resulted in a marked increase in blood flow of the treated hand in three of the four subjects studied. Maximum flows amounting to as much as twenty times the control flow were observed. The augmentation in flow usually occurred within fifteen minutes after the iontophoresis began, reaching a maximum within thirty minutes. Systemic effects were negligible or absent, and no changes in skin temperature or blood flow of the untreated hand were noted. No effect resulted from ABMC without current, or from iontophoresis of normal saline. The ability of ABMC to overcome locally the vasoconstriction produced by cold may be of major significance in studies of peripheral vascular adjustments to environmental temperature and warrants further investigation.

Responses to small changes of light intensity by the light-adapted photoreceptor. E. F. MACNICHOL (by invitation) and H. K. HARTLINE. *Johnson Research Foundation, University of Pennsylvania, Philadelphia.* Action potentials were recorded from single optic nerve fibers from the eye of *Limulus*. The preparation was subjected to a constant illumination lasting for several seconds. When the discharge of nerve impulses reached a constant frequency the illumination was changed by small increments and the time course of the change in the frequency of discharge was followed. In response to a small increase in illumination a large transient increase in frequency was produced. The frequency eventually subsided to a steady level slightly greater than that preceding the change in illumination. A small decrease in illumination was likewise followed by a large decrease in frequency followed by a gradual recovery to a level slightly below that previous to the change in illumination. If a flash of light lasting several seconds was added to a steady background of illumination the transient at the beginning and end were nearly but not exactly symmetrical; the number of impulses added at the onset of the flash was very nearly equal to the number of impulses missed at its termination. The magnitude of the transient change in frequency depended on the level of background intensity and on the magnitude of its increment, and was approximately constant for a fixed incremental ratio. Duration of the intensity increment and of the foregoing period of steady background illumination affected the magnitude of the transient.

Antigonadotrophin formation to sheep FSH: effectiveness against endogenous gonadotrophic hormones in men. WILLIAM O. MADDOCK, EDWIN C. JUNGCK (by invitation), and Carl G. HELLER. *Department of Physiology, University of Oregon Medical School, Portland, Oregon.* Seven infertile men, each having some functional testicular tissue and detectible amounts of urinary gonadotrophins (with one exception) were given intramuscularly 50 to 100 rat units of a purified follicle-stimulating

hormone prepared from sheep anterior pituitaries (Schering's FSH, containing small amounts of ICSH and thyrotrophin) daily for 60 to 105 days. Anti-gonadotrophins, although not demonstrable before treatment, appeared in the plasma from 49 to 60 days after beginning therapy. These titers were sufficiently great to nullify the effect of 100 times the amount of FSH administered. Highest titers were reached at the time of stopping therapy. At this time 0.9 cc. of plasma completely inhibited the effect of 2 rat units of FSH administered to immature rats (usually 2 rat units of FSH caused an increase in ovarian weights from circa 13 to circa 30 mgm.). Four to five months after cessation of therapy anti-gonadotrophin titers were still approximately half as high as peak levels. Injecting FSH did not cause a rise in urinary gonadotrophin output. Detectable amounts of urinary gonadotrophin were found at the time of maximal anti-gonadotrophin formation.

The patient's plasma anti-gonadotrophin capacity was tested against the patient's own urinary gonadotrophins in several instances, and also against other patients' urinary gonadotrophins, and were effective against both. Therefore, either the patients' kidneys or the ultrafiltration concentration method separated endogenous gonadotrophin from endogenous anti-gonadotrophin. This indicates that anti-gonadotrophins do not destroy, inactivate or irreversibly combine with gonadotrophins.

Effect of hydrogen and bicarbonate ions on the metabolic rate of the perfused cat brain. J. MAGNES and A. GEIGER (introduced by H. GRUNDFEST). *Department of Physiology, the Hebrew University, Jerusalem and Department of Psychiatry, College of Physicians and Surgeons, Columbia University.* Isolation of the cerebral circulation and perfusion of the brain in the living cat, by the method described previously (A. Geiger and J. Magnes, Amer. Journ. of Physiol. **149:** 517, (1947)), offers an opportunity to study the effect of blood composition on the metabolic functions of the brain. The effect of (H^+) and of HCO_3^-) was studied while perfusing the brain with defibrinated heparinised ox blood. Acidifying the blood with HCl or with lactic acid increased cerebral oxygen consumption considerably; for instance, a change from pH 7.25 to pH 7.20 usually caused an increase of 25–30% in oxygen consumption. Addition of $NAHCO_3$ caused a corresponding depression. On the other hand comparable changes in hydrogen ion concentrations, brought about by varying the CO_2 tension of the perfusing blood were inconsistent in their effect on brain metabolism and were often without any influence whatsoever. The metabolic rate of the brain is highest when perfused with blood having a low $NaHCO_3$ concentration. (This was obtained by dialysing blood against 0.9% NaCl, and subsequently adding all the electrolytes and glucose in physiological concentrations with the exception of bicarbonate, which was kept low.) These experiments suggest that hydrogen ions stimulate, and bicarbonate ions depress, brain metabolism.

Urea-clearance by perfusion of the entire intact small intestine in man. N. S. R. MALUF. *Department of Pharmacology, University of Louisville School of Medicine, Louisville, Ky., and the Louisville General Hospital.* Perfusion was through a modified Miller-Abbott tube. The 300 cm.-long, 16 French modified tube has three conduits: 1. for inflating the terminal balloon when the tube has entered the duodenum; 2. for passing the perfusing liquid into the proximal jejunum through 5 holes about 150 cm. (5 ft.) from the distal end; 3. for sucking back the liquid at the distal ileum through 5 holes immediately proximal to the balloon. The balloon is deflated when the distal end of the tube is in the lower ileum. The solution was somewhat hypertonic Na_2SO_4. Suction was set to read −5 to −10 mm. Hg at junction of intestinal tube and tubing connected to reservoir and suction-pump. With the rate of perfusion at 30 cc./min., the concentration of urea in the outflow liquid was almost identical with that in the plasma, giving a urea-loss of 11.6 Gm. in 8 hours, or a urea-clearance of 32.4 cc./min. This compares well with peritoneal irrigation and external dialysis ("artificial kidney"). The method is much less cumbersome and requires no asepsis or surgery; furthermore, it may be temporarily discontinued when desired. The method is indicated in temporary acute renal insufficiency, as after transfusions with incompatible blood, sulfonamides, or in crush-syndrome. The chloride-loss in 8 hours was 6.64 Gm. (as NaCl); this can be readily replaced by NaCl by intravenous drip. As shown by Amberson, SO_4 may replace Cl to a large extent without observable injury.

The platelets as foci in the coagulation of the blood. F. D. MANN, MARGARET HURN and D. R. MATHIESON (introduced by T. B. MAGATH). *Division of Clinical Laboratories, Mayo Clinic, Rochester, Minnesota.* The coagulation of normal human plasma in silicone-coated slide preparations was observed by phase contrast microscopy. As soon as brin threads were visible they were seen radiating from the platelets. Thus the platelets appeared to serve as foci of fibrin formation, but simultaneous formation of fibrin free in the plasma is not excluded. After coagulation the platelets were firmly and for the most part individually fixed in the fibrin network and did not migrate. The relative prothrombin-converting activities of plasma and of platelets were studied with the aid of aged, zymin-treated and ammonia-treated plasma. The ability to restore the coagulability of aged and ammonia-treated plasma was greatly concentrated in or on the platelets. This agrees with previous evidence

indicating at least two components in the prothrombin conversion factor. The concentration of prothrombin conversion factor in or on the platelets appears to be related to the function of platelets as foci in coagulation of the blood.

The separate contraction of functional areas in limb muscles during reflex withdrawal and crossed extension. J. E. MARKEE and MAUDE WILLIAMS (by invitation). *Dept. of Anatomy, Duke University School of Medicine, Durham, North Carolina and Division of Physiology, Womens College of U. N. C., Greensboro, North Carolina.* Previously we have demonstrated in the dog that the separately innervated areas within a muscle may contract separately by electrical stimulation of their nerves, and in the human, that the arrangement of the nerve branches to the long limb muscles is similar. In this study we recorded, photographically and electrically, the shortening of separately innervated areas under reflex conditions—ipsilateral withdrawal and crossed extention—of all the limb muscles which move both hip and knee joints. Sixty-five decerebrate and spinal dogs were used.

The predominate reflex response of the long thigh muscles is as follows: In the semitendinosus the proximal part shortens during extension of the hip; the distal part, during flexion of the knee. The semimembranosus shortens during flexion of the knee only. The lateral part of the biceps femoris shortens during knee flexion; the medial, during hip extension. The caudal part of the sartorius shortens only during knee flexion; the cephalid cranial part during hip flexion; the distal cranial part during knee extension. The superficial part of the rectus femoris shortens during knee extension; the deeper part, during hip flexion. The superior part of the gracilis shortens during knee flexion; the inferior part, during hip extension. Certain different functional areas of a muscle, under reflex stimulation, react as if reciprocally innervated. Action potentials were recorded in areas that shorten, but not in areas that lengthen.

Purification of renin. JOHN MARSHALL (by invitation) and G. E. WAKERLIN. *Department of Physiology, University of Illinois College of Medicine, Chicago.* A modification of earlier methods for the purification of renin was developed for the purpose of producing large amounts of relatively pure hog renin in high yields. Dried hog kidney powder prepared by acetone-ether extraction was used in lots of 10 kg. of fresh kidney equivalent. Each lot was extracted with saline-bicarbonate at a pH of 8. The extract was filtered through muslin, and inert protein precipitated at pH 2.90 at an NaCl concentration of 0.92 M., using HCl (2N) to pH 4 and trichloracetic acid (10%) to pH 2.90. After centrifugation the supernatant was saturated with NaCl and the precipitate of crude renin was separated by centrifugation. This was then dialyzed until essentially salt free. Total volume was 1 liter, containing 16 Goldblatt renin units per ml., at a purity of 8 units per mg. N. Further purification was achieved by ethanol fractionation under controlled conditions of ethanol concentration, pH, ionic strength, temperature, and protein concentration. Inert protein was removed at pH 7.0, EtOH 17%, $\gamma/2$ 0.01 to 0.003, T $-5°$. This precipitate consisted of 10–15% of the total protein and 10% of the total renin. (Increasing ionic strength or pH yielded less protein, and increasing ethanol to 20% precipitated 60% of the renin.) After centrifugation at $-5°$, the supernatant was adjusted to pH 4.90 with 0.8 M acetate buffer. The precipitate formed contained about 15% of the initial total protein, and 80–90% of the total renin at a purity of 50 units per mg. N. Further purification with minimal loss of renin was achieved by a second fractionation at lower ionic strength and ethanol concentrations. This work is continuing.

The rate of atrophy of rat diaphragm. ARTHUR W. MARTIN and O. M. SOLA (by invitation). *Department of Physiology and Biophysics, University of Washington, Seattle, Washington.* The rate of muscle atrophy varies approximately inversely with increasing animal size. While it has not been possible to relate the rate of atrophy directly to the rate of oxygen consumption Diaz-Guerrero and Thomson (Fed. Proc. **6:** 97, 1947) showed that thyroxin administration increased and thiouracil decreased the rate of atrophy of muscle. It appeared of interest to compare the rate of atrophy of a muscle with a high metabolic rate with one of low metabolic rate in the same species of animal. Field, et al. (J. Cell. Comp. Physiol. **14,** 143, 1939) showed that rat diaphragm consumed oxygen at about twice the rate of other skeletal muscles. Abundant data have been obtained on the rate of atrophy of other muscles of the white rat, particularly following denervation. We have therefore determined the rate of atrophy of a hemi-diaphragm in this species following section of the left phrenic nerve in the thorax. Seven days after nerve section a maximum hypertrophy of 50 per cent had taken place, although by this time gastrocnemius muscle would have atrophied about 23 per cent. The hypertrophy was gone after 30 days by which time gastrocnemius atrophy would be about completed (75 per cent). Progressive atrophy followed, a 25 per cent weitht loss occurred by 70 days, a 50 per cent loss by 120 days.

No significant changes in absolute amount of connective tissue took place within 120 days. Even when allowance was made for the amount of connective tissue the QO_2 was significantly elevated (22 per cent) during hypertrophy and reduced during atrophy.

The effects of various pressure application de-

vices (anti-g suits) upon arterial and venous pressures. E. M. MARTIN (by invitation), M. J. MARTIN (by invitation), J. P. HENRY, J. L. GAMBLE, JR. (by invitation) and R. S. SHAW (by invitation). *Aero Medical Laby., Air Meteriel Command, Wright Field, Dayton, Ohio.* The anti-g suit is a convenient devide for suddenly compressing an extensive area of the body and driving the blood contained therein into the rest of the circulatory system. In the work reported, three different anti-g suits were employed: the G-3A, G-4A, and the modified Clark-Navy capstan suit. They were designed to compress the lower limbs and abdomen with progressively greater efficiency in the order mentioned, G-3A, G-4A, and the capstan anti-g suit. In an attempt to analyze the mechanism whereby the suit protects against the effects of positive g, venous and arterial pressures were studied both at rest and under g. In ten tests on five subjects, the relative efficiency of rhythmic inflation of the G-3A and G-4A suits in reducing venous pressure in the foot when sitting still at 1 g was compared. There was no significant difference in pressure reduction, i.e., G-3A, 60%; G-4A, 55% of resting pressure. This result suggests that there is no difference in their efficiency in returning blood from the legs under these conditions. However, their efficiency in elevating mean arterial pressure both as measured when at rest using the sphygmomanometer and under g using the radial artery appears to be dependent on the above-mentioned efficiency and uniformity with which they compress the lower half of the body.

The projection of area 19 to the reticular formation. W. S. MCCULLOCH and ELWOOD HENNEMAN. *Department of Psychiatry, University of Illinois.* After local strychninization of area 19 in the cat under Dial, its brain stem was explored with 6 parallel micro-electrodes, spaced with points 2 mm. apart, carried on a stereotaxic apparatus. 5 channels of a Grass electroencephalograph were used to record from these needles in push-pull arrangement between adjacent electrodes. The 6th channel recorded from area 19. No strychnine spikes were found except in the medial reticular suppressor formation of the brain stem. It is, therefore, concluded that area 19, like areas 4S and 24 fires into the suppressor part of the reticular formation.

A critical discussion of the bioelectric "doublet" theory. RICHARD MCFEE (introduced by J. S. Robb). *Department of Pharmacology, Syracuse University College of Medicine.* The mathematical derivation of the "doublet" theory contains an inconsistency. The determination of the potential field of a polarized membrane, fundamental to this theory, assumes a homogenous surrounding medium. However, the membrane specific resistance is very much greater than that of the fluid conductor and consequently the medium is non-homogenous. The effect of the non-homogeneity can be determined by measuring the potential at points about an electrostatis equivalent to a partly depolarized fibre. Such measurements show a potential field very different from that of a doublet. The effect of this field on the difference of potential between local and remote electrodes can also be measured and plotted as a function of the degree (or position) of the depolarization of the simulated fibre. This results in polyphasic waves, due to variations at both local and remote electrodes. Furthermore, the potential at the remote electrode is dependent only on the state of depolarization of the fibre and not the distance from it. This suggests that the "doublet" theory derived concept of the "indifferent" electrode be seriously questioned.

Although no data on living single fibre preparations has been obtained at this laboratory an experiment by Lorente de No, on potentials produced by action currents of an isolated nerve fibre in a plane conductor (Howells Textbook of Physiology, 15th Ed.), indicates good agreement with local-remote potential curves determined from the equivalent fibre constructed here.

Spinal projection of hind-limb afferent fibers. A. K. MCINTYRE (by invitation) and David P. C. LLOYD. *Laboratories of The Rockefeller Institute for Medical Research, New York.* Velocity, in dorsal roots and columns, of impulses initiated in the largest afferent fibers of muscle (Group I) and skin (Group II) has been measured, utilizing gastrocnemius (L_7, S_1) and sural nerves connecting with lower hind-limb segments, and quadriceps ($L_{4,5,6}$) and saphenous nerves connecting with upper hind-limb segments. *Muscle afferent fibers* exhibit characteristic nerve-root velocities to the upper limits of their root entry, at which points velocity falls to about 60%. Further velocity decreases occur, notably in L_3, until, in lowest thoracic segments it is about 30%. No trace of Group I impulses has been found above T_{10}, although muscle Group II has been traced to uppermost thoracic segments. *Cutaneous afferent fiber* velocity change is less marked, impulses projecting through the dorsal columns, at approximately 75% nerve velocity until, in upper thoracic segments, further decrease occurs. At T_{12} or L_1 these impulses outstrip the initially earlier muscle Group I impulses. Antidromic column stimulation, recording at several points on the nerves, yields similar results, and permits, in addition, proof of nerve velocity of the impulses. By this means also it can be shown that muscle Group II impulses pass the initially faster Group I impulses in the lower thoracic cord to continue upward. Amplitude of ascending Group I volleys decreases in two distinct stages, the discontinuity occurring uniformly in L_3. These data are interpreted to mean that Group I fibers terminate by supplying cells of

the lower thoracic cord and upper three lumbar segments, presumably Clarke's column, whereas Group II fibers, of muscle and cutaneous origin, contribute to the tract of Goll.

On estimating the relative volumes of corpuscles and serum in blood. PAUL L. MCLAIN and C. H. WILLIAM RUHE (introduced by C. C. GUTHRIE). *Department of Physiology and Pharmacology, School of Medicine, University of Pittsburgh.* Relative corpuscle and serum volumes were estimated in numerous samples of defibrinated beef blood by 12 technically different applications of the serum dilution principle. The results were compared with those obtained on the same samples by centrifugalization at approximately 7500 x G. In the dilution studies, the following measurements on serum were used as indices: specific gravity by copper sulfate and falling drop methods; total nitrogen; serum protein estimated from nitrogen determination and from specific gravity; heat-coagulable serum solids by direct weighing and from specific gravity; concentration of the dye T-1824. With regard to relative corpuscle volume, the means for all dilution methods were smaller than that for the conventional hematocrit, differences varying with the method from 1 to 20 per cent of the mean cell volume. The average differences, for comparisons on individual samples, between centrifugal and dilution procedures covered a similar range. In general, the dilution methods which gave the smallest deviations from the centrifugal, especially those which involved washing the serum from the packed corpuscles, were the most consistent. However, present results do not justify the selection of any one method as estimating reliably the "true" cell volume, or the practice of correcting conventional hematocrit results by the application of any constant factor based on dilution methods.

Preliminary study of complete parenteral alimentation. H. C. MENG (by invitation), and SMITH FREEMAN. *Department of Physiology and Department of Experimental Medicine, School of Medicine, Northwestern University, Chicago, Illinois.* Two adult male dogs were fed a fat-free synthetic diet by mouth plus 10% butter oil emulsion by vein for 4 weeks, after which the animals were infused exclusively through the intravenous route with mixtures containing all the nutritional elements in adequate amounts except water which was allowed by mouth *ad libitum*. Two types of mixture were used. One contained 10% butter oil in emulsion with the stabilizers—span 20, "Asolecten" and sodium cholate—and 10% glucose. The other contained a 10% casein hydrolysate ("Amigen"), glucose, salt mixture and water soluble vitamins. Fat soluble vitamins and liver extract were injected intramuscularly every week. The total daily caloric intake was 92.65 calories per kilogram of which 49% came from carbohydrate, 19% from protein and 32% from fat. On some occasions the butter oil emulsion was omitted and the calories from the fat were made up by glucose. The animals were infused for 8 and 10 weeks. There was no change in total plasma proteins, plasma N.P.N., Rose Bengal clearance and serum phosphatase. Their body weight was apparently maintained. The nitrogen balance was positive except on 1 or 2 occasions. No pyrogenic effects were observed. Vomiting occurred when infusion was given too rapidly. A moderate reduction of hemoglobin, red cell count and hematocrit was noticed after the first week of injection, but did not progress appreciably thereafter. Hematuria occurred on several occasions. Further studies are in progress.

Removal of rostral border of human area 4 followed by spasticity or lack of it. FRED A. METTLER and J. LAWRENCE POOL (by invitation). *Department of Neurology, College of Physicians and Surgeons, Columbia University.* Hines and Tower showed that removal of the caudal part of the simian area gigantopyramidalis produced no spasticity and imputed such spasticity as does follow removal of area 4 to damage of its rostral border. Mettler (1943, J. comp. Neurol. **79:** 185-245) demonstrated that unilateral decortication produces negligible spasticity but that progressively deeper ablations ultimately produce the complete hemiplegic picture. Four human cases are here presented. In case K, area 6 (except caudal border) and rostrally adjoining cortex was unilaterally removed. Stimulation still gave good movements. Postoperative paralysis with resistance to passive movement developed. In case M the caudal third of the middle frontal gyrus was unilaterally removed without result. The precentral and part of the postcentral gyri caudal to the original ablation were next removed. A flaccid paralysis developed from which the patient recovered without showing evidence of spasticity. In cases E and Y, area 6 was bilaterally removed without eliciting any evidence of spasticity. Subsequently the rostral part of area 4 was removed unilaterally in both cases. In case Y a pronounced spastic hemiplegia developed, in Case E a flaccid paralysis ensued. Since cases K and Y had pre-existing subcortical pathology and cases M and E did not, it is concluded that unilateral removal of neither area 6, nor of 6 and the rostral part of 4, nor of 6 and both the rostral and caudal parts of 4 (and the rostral part of the postcentral gyrus) produces notable spasticity in the human unless subcortical damage also exists.

Effect of delta- and gamma- tocopherol on creatinuria in progressive muscular dystrophy. A. T. MILHORAT. (*Depts. of Psychiatry and Medicine, Cornell Univ. Medical College, The Russell Sage Institute of Pathology and the New York Hospital, New York, N. Y.*) Investigations reported previously (Milhorat and Bartels, Federation Proc. **6:**

414, 1947) showed that certain sugars present in gastric mucin and pectin can increase the utilization of alpha-tocopherol in progressive muscular dystrophy. Alpha-tocopherol alone was without effect on creatinuria; when given together with D-Mannose, D-Galactose or L-Fucose alpha-tocopherol induced pronounced reduction in the creatine output of suitable subjects. Through the kindness of Dr. James G. Baxter of Distillation Products, Inc., Rochester, N. Y. who supplied us with samples, we have had the opportunity of testing the effects of gamma-tocopherol and delta-tocopherol. The patients who were given test doses of these substances were maintained on a constant creatine-free diet and had been used in the earlier investigations of alpha-tocopherol. Gamma-tocopherol administered in daily doses of 330 mg for 3 days either alone or with D-Mannose or raffinose was without effect on creatinuria. On the other hand, delta-tocopherol given for similar test periods in amounts of 220 mg. daily reduced the creatinuria as much as 40 per cent for 9 days in some patients. The effect was greatest in patients with benign forms of the facial-humeral scapular type of muscular type of muscular dystrophy, and less, or even absent, under these conditions, in patients with rapidly progressive forms of the pseudo-hypertrophic type of the disease.

The influence of oxygen administration on the cardiovascular response to exercise. A. T. MILLER, JR. *Laboratory of Applied Physiology, University of North Carolina, Chapel Hill.* Three subjects, thoroughly trained in treadmill running, performed standard exercise which consisted in running on the treadmill (speed 7 m.p.h., grade 17.5 per cent) for 5 minutes. Heart rate and systolic blood pressure were recorded at one minute intervals during exercise and 20 minutes recovery. Blood lactate concentration was determined before and after exercise and after 20 and 60 minutes recovery. Oxygen was administered for 10 minutes before exercise, during exercise, during a 20 minute recovery period and during a 5 minute interval between two identical exercise periods in different series. Each series consisted of 3 or more experiments on each of the 3 subjects, with an equal number of control experiments in which air was breathed.

Two of the subjects made repeated exhaustion runs (3 minutes at 12 m.p.h., 17.5 per cent grade) in addition to the standard runs. The results may be summarized as follows: (1) Oxygen administration *before* exercise had no effect on heart rate, blood pressure and blood lactate during exercise and recovery. (2) Oxygen administration *during* exercise had no effect on heart rate or blood pressure during exercise or recovery. Maximal blood lactate concentration was reduced to a slight but statistically significant extent (confirmed by experiments on 20 additional subjects). (3) Oxygen administration *during recovery* had no effect on the rate at which heart rate, blood pressure and blood lactate returned to normal. (4) Oxygen administration between double runs had no effect on heart rate, blood pressure and blood lactate during the second run and recovery.

Insulin excretion in normal man. I. ARTHUR MIRSKY, JOHN WACHMAN (by invitation), C. J. PODORE (by invitation) and R. H. BROH-KAHN (by invitation). *May Institute for Medical Research of the Jewish Hospital, Cincinnati, Ohio.* The amounts of insulin excreted into the urine by normal subjects were determined by the following procedure. Several days' urine from each subject was desiccated by lyophilization. The resultant powder was then extracted by the usual acid alcohol procedure and the insulin recovered by precipitation with ether. The insulin content of the final acid saline extract was determined by the Young modification of the mouse dropping test. Recovery experiments indicated that from 60 to 100 per cent of 0.5 unit of insulin added to large volumes of urine could be detected by this procedure. Numerous trials have indicated that under ordinary conditions of diet, normal subjects excrete very small amounts of insulin into the urine. The average amount of insulin determined in a series of such normal subjects was found to be about 0.3 units per day. Further trials have indicated that, even after the injection of fairly large amounts of insuln into normal subjects, little additional insulin appeared in the urine. This suggests that normal man excretes only minute mounts of insulin into the urine. In view of the probability that exogenous insulin rapidly disappears from the blood stream of man, these findings would be indicative of the very rapid rate of destruction of insulin by the intact organism.

Calibration of the Millikan compensated oximeter as used among white and colored persons. GEORGE E. MONTGOMERY, JR (by invitation), J. E. GERACI (by invitation) and EARL H. WOOD. *Section on Physiology of the Mayo Clinic and Foundation, Rochester, Minnesota.* The Millikan CMR model no. 13 compensated circuit oximeter has been calibrated against Van Slyke analyses of simultaneously drawn samples of arterial blood from twenty-five normal white persons and five normal Negro persons. Multiple samples of arterial blood were obtained by the technic described by Wood and associates (see abstract). Hypoxia was produced by allowing the subjects to breathe gas mixtures of from 16 to 7 per cent oxygen content. Ten different Coleman-type earpieces were used. Five hundred fourteen simultaneous oximeter and Van Slyke determinations of arterial oxygen saturation were made in the group of twenty-five white persons In the saturation range from 50 to 91 per cent, the

average difference between the oximeter and the Van Slyke values was +4.7 percentage points (range, −11 to +23 percentage points). The accuracy of the oximeter was found to vary significantly when different earpieces were used. The average differences between photo-electrically and chemically determined arterial oxygen saturation values ranged from +1.4 to +13.1 percentage points for the individual earpieces tested. Sixty-two comparisons of oximeter-determined and Van Slyke-determined values of the arterial oxygen saturation were made in a group of five Negro and five white persons, the same oximeter earpiece being used. In the 50 to 90 per cent saturation range in Negroes, the average difference between Van Slyke and oximeter values was +12.8 (1.7 to 22.7) percentage points. In the white persons this value was +7.1 (−1.1 to 15.7) percentage points. Figures in parentheses are extreme values.

Estimation of the total circulating red cell volume by the use of methemoglobin-tagged cells. JAMES C. MOORE and O. W. SHADLE (introduced by HAMPDEN C. LAWSON). *The Dept. of Physiology, Univ. of Louisville School of Medicine.* Sodium nitrite was added to heparinized blood in the proportion 50 mgm./100 cc. for *in vitro* methemoglobin formation. The methemoglobin-containing cells were washed with saline and injected intravenously as concentrated cell suspensions in volumes of 35–70 cc. in barbitalized, splenectomized dogs. Methemoglobin was determined spectrophotometrically by the method of Horecker and Brackett on arterial samples drawn at intervals of one to five minutes after the injection. Methemoglobin disappearance curves similar to those found *in vitro* were obtained after the first five minutes. Arterial methemoglobin at the instant of injection was obtained by extrapolation of these curves. Cell volume was calculated from the total methemoglobin injected and the packed-cell methemoglobin concentration in arterial blood, both values being read at the instant of injection, the latter by extrapolation as above. *In vitro*, this method gave values for cell volume within 2 per cent of those obtained from the hematocrit and the total volume.

In a series of six dogs comparative values for cell volume were obtained by the present method, by calculation from the hematocrit and the plasma volume, and by the "hematocrit-change" method of Lawson, Overbey, Shadle, and Moore. The second method gave values ranging from 126 to 152 per cent, and the third method 91 to 130 per cent of the volumes obtained by the present method.

Studies of shock in frogs. 1. Shock due to electrical injury. LOUIS MOREAU (by invitation), MARVIN BALISTOCKY (by invitation), and L. V. HEILBRUNN. *Zoological Laboratory, University of Pennsylvania.* The frog is an excellent animal in which to study shock phenomena. Some of the complications which enter into the shock picture in mammals are not present in the frog. We have found that shock may be caused by electrical injury to a restricted region of the animal. If either the brain of the legs are subjected to currents sufficiently strong to cause injury (but not excessive heat) the animals show obvious shock. Toxic factor is involved, for ligature of the legs prevents shock in frogs whose legs are exposed to electrical injury. The toxic factor is apparently of the nature of a thrombin or thromboplastin. This is indicated by the changes which occur in the coagulation time of the blood of shocked animals. The first change is a slight decrease in the coagulation time and this is followed by an increase.

The effect of intraarterial injection of acetylcholine upon the gastric mucosa of the dog. G. M. MORTON (by invitation) and G. W. STAVRAKY. *Departments of Anatomy and Physiology, Faculty of Medicine, University of Western Ontario, London, Canada.* As reported by Stavraky (Federat. Proc. 4: 1, 1945) acetylcholine, when injected into the gastrosplenic artery, causes a secretion of alkaline gastric juice. A histologic study of the gastric mucosa of the injected region of the stomach—a limited area of the body along the greater curvature—showed that after 4–6 hours of injection of acetylcholine, complete exhaustion of the chief cells of the neck of the gastric glands and partial exhaustion of the surface epithelium took place. In contrast to the quiescent areas of the stomach, when stained with mucicarmine, the chief cells of the neck of the glands contained no secretory granules, their cytoplasm appearing quite clear. The cells of the surface epithelium, particularly those in the depths of the gastric pits, contained diminished amounts of the mucus precursor, the secretory granules occupying only the outer regions of the cells and leaving a clear area between the mass of the granules and the nucleus. When stained with neutral gentian, the chief cells of the body of the glands were found to contain large numbers of pepsinogen granules showing no difference between the injected and resting mucosa. This latter was in keeping with the absence of any digestive power in the secreted juice.

It is concluded that the highly alkaline gastric juice (pH up to 8.9) which is secreted in response to injections of acetylcholine into the gastrosplenic artery comes from the inner one-third of the glands of the body of the stomach along the greater curvature, the chief cells of the neck and possibly the surface epithelium being responsible for its secretion.

The role of the nervous system in experimental hypertension in the dog. W. G. Moss (by invitation) and G. E. WAKERLIN. *Department of Physiology, University of Illinois College of Medicine.* A variety of techniques for the study of the role of

the nervous system have been employed in an analysis of its role in the maintenance of the blood pressure level in experimental renal hypertension. The results bear out the following conclusions. The applications of various drugs which produce surgical anesthesia in dogs demonstrates by the changes in pressure levels that there is no quantitative difference in the role of the nervous system in the maintenance of the blood pressure level in normotensive, early renal hypertensive, or late renal hypertensive dogs. There is a large increase in the role of the nervous system in the maintenance of the pressure level in neurogenic hypertensive dogs. Blocking the vasomotor outflow between the central nervous system and the blood vessels causes changes in the blood pressures of dogs which also demonstrate no increased role for the nervous system in the maintenance of the blood pressure level in renal hypertensive dogs in either early or late stages. Neurogenic hypertension is again shown to involve an increased role of the nervous system. Studies of reflex mechanisms in normotension and late renal hypertension show little difference in the responses to hypercapnic and cold stimulation. The responses of early renal hypertensives to these stimuli are somewhat less. Insofar as these experiments are concerned, the role of the nervous system in the maintenance of the blood pressure level in experimental renal hypertensive dogs does not appear to differ from its role in normotensive dogs.

A polarographic study of the action of carbonic anhydrase at the dropping mercury electrode. Otto H. Müller. *Dept. of Physiology, Syracuse University College of Medicine, Syracuse, N. Y.* In a polarographic study of quinhydrone in buffered and unbuffered solutions it was found that the carbon dioxide-bicarbonate buffer behaved abnormally (Müller, J. Am. Chem. Soc., 1940, **62:** 2434). Instead of a single wave in which the anodic and cathodic portions overlap, two waves were obtained which were similar yet not identical with those found in an unbuffered solution. This was attributed to a slowness in the response of the buffer to changes in pH at the electrode surface. If carbonic anhydrase (prepared from sheep erythrocytes by the customary alcohol-chloroform method) is present in sufficient concentration and all inhibitors (such as NO_3^-, Cl^-, etc.) are absent, the carbon dioxide-bicarbonate buffer reacts with sufficient rapidity to maintain constant the pH at the dropping mercury electrode during the reduction or oxidation of quinhydrone. This is evident from the fact that the anodic and cathodic waves now overlap completely and form a single wave, to separate again more or less as the enzyme is destroyed or inhibited by well-known inhibitors.

Composition of Central American foods. I. Honduras. Hazel E. Munsell, Robert S. Harris and Louis O. Williams (by invitation), with the technical assistance of Louise Guild (by invitation), Gertrude Nightingale (by invitation) and Cynthia Troescher (by invitation). *Nutritional Biochemistry Laboratories, Massachusetts Institute of Technology.* For the establishment of a sound program of food production and food consumption it is important to know the composition of the indigenous foods which are, or can be, grown in the area. On this basis a long range study of the composition of foods grown in Central America is underway. Specimens are collected and identified by a botanist, samples representative of the edible portion are stabilized and shipped by air express to Cambridge. Whenever possible a record is made of the soil type, annual rainfall, altitude and whether the sample was grown of fertilized land. A comparison of the data from similar samples may give information on the relative importance of genetic and non-genetic factors in determining the composition of edible plants. Moisture, nitrogen, ether soluble fraction, crude fiber, ash, calcium, phosphorus, iron, carotene, thiamine, riboflavin, niacin and total ascorbic acid content are being measured. In the present report the results of analyses of 129 samples of 85 plants collected in Honduras are presented.

The effects of the intravenous administration of glucose and amino acids on the hepatic blood flow and splanchnic oxygen consumption of man. By J. D. Myers (Introduced by E. A. Stead, Jr.), *Dept. of Medicine, Duke University, School of Medicine, Durham, N. C.* The work of Wilhelmj, Bollman and Mann on the dog, and of Dock on the rat, has indicated that the specific dynamic action of proteins and amino acids takes place chiefly, if not solely, in the liver. Mann and Boothby have established that in the dog the specific dynamic action of glucose is not only still present after hepatectomy, but is actually accentuated in the absence of the liver. The estimation of hepatic blood flow by bromosulphalein clearance during catheterization of the hepatic veins, and the calculation of the splanchnic oxygen consumption (the estimated liver blood flow × the hepatic arterio-venous oxygen difference) have provided means of investigating the immediate effects of the intravenous administration of amino acids and glucose on the hepatic blood flow and splanchnic oxygen consumption in a group of hospital patients without significant disease. The rapid intravenous injection of 50 grams of a mixture of amino acids in 10 per cent solution (Vujn 9 solution—Merck) has resulted during the subsequent hour in an increase in splanchnic oxygen consumption to as much as 250 per cent. This is accomplished by a marked increase in hepatic arterio-venous oxygen difference with no significant change in hepatic blood flow. At the same time, there is an outpouring of glucose from the liver as evidenced by an increased hepatic venous-arterial glucose difference. The similar in-

jection of 50 grams of glucose in 10 per cent solution has resulted in a decided fall in splanchnic oxygen consumption due to a decrease both in hepatic blood flow and in hepatic arterio-venous oxygen difference. The marked hyperglycemia attendant upon the rapid injection of glucose was consistently accompanied by an immediate and marked increase in hepatic venous-arterial glucose difference indicating a liberation of glucose from the liver. The influence of epinephrine and insulin on the changes found after glucose administration is presented. These observations indicate that in man, and in agreement with the observations in lower animals, the increased oxygen consumption following the intake of amino acids takes place in the splanchnic area, and presumably in the liver. The increased oxygen utilization after glucose administration occurs elsewhere than in the splanchnic area. In fact, the decreased oxygen consumption in the latter viscera tends to mask the increase which occurs elsewhere.

The difference between pharmacological action and the physiological role of acetylcholine. DAVID NACHMANSOHN and MORTIMER A. ROTHENBERG (by invitation). (*Department of Neurology, College of Physicians & Surgeons, Columbia University, New York.*) The theory of chemical mediation of the nervous impulse was based on the pharmacological action of acetylcholine limited to synapses and neuromuscular junctions. Failure of massive doses of acetylcholine (20 grams per liter) to affect nerve fibers was considered a proof that acetylcholine has no physiological role in conduction. Studies on the role of acetylcholine in nervous function with methods of biochemistry and biophysics have conclusively established that the release and the removal of acetylcholine are essential events in conduction. It has been demonstrated that the failure to affect fibers exposed to acetylcholine has to be attributed to the inability of the ester to penetrate the lipoid membrane. Acetylcholine, like prostigmine and curare, is a methylated quaternary ammonium salt. It could be shown that prostigmine, in contrast to di-isopropyl fluorophosphate and eserine, a tertiary amine, does not affect the action potential and does not penetrate into the interior. More recently, direct evidence was offered by the use of the ester labelled with N^{15}, that the nerve membranes are impervious to acetylcholine (Rothenberg, Sprinson & Nachmansohn). The experiments show that the pharmacological effect of acetylcholine, which was the basis of the transmitter theory, does not permit conclusions as to the site of physiological action. Effects of compounds applied externally depend on many variables. The investigations with anticholinesterases offer an excellent illustration how rate of penetration, affinity to the enzyme, chemical constitution, and other factors to be discussed, influence the effect of compounds to which the intact nerve fiber is exposed.

The nature of unipolar limb leads in the dog. L. H. NAHUM, H. M. CHERNOFF (by invitation), and W. KAUFMAN (by invitation). *Laby. of Physiology, Yale Univ. School of Medicine, New Haven, Conn.* Electrocardiographic exploration of the dog's heart by five different methods (impressed potentials, forced extrasystoles, epicardial KCl application, coronary artery ligation, and heating and cooling the epicardium) permitted the determination of the ventricular regions represented in Leads Vr, Vl, and Vf. The ventricles could be divided into three zones: a) *proximal*, facing the exploring electrode; b) *distal*, facing away from the exploring electrode; c) *intermediate*, a narrow region separating the proximal and distal zones. The distribution of these zones for each lead will be described elsewhere. Depolarization within the proximal zone of a selected unipolar limb lead caused a downward deflection in the complex in that lead, depolarization within the distal zone produced an upward deflection; depolarization within the intermediate zone is not detected in the exploring lead. Accelerating repolarization by heating the proximal zone increased T-wave positivity; decelerating depolarization by cooling the proximal zone produced increased T-wave negativity. Heating and cooling distal zones produced opposite effects. The electrocardiogram in each unipolar limb lead in the dog is derived from the algebraic summation of simultaneous electrical events taking place in proximal, intermediate, and distal ventricular zones. These experiments lend no support to the view that unipolar leads are derived from an endocardial-epicardial interference.

The sodium chloride tolerance of female mice. DOROTHY NELSON (Introduced by JOHN S. GRAY). *Physiology Dept., Northwestern University Medical School.* Mammals, including the mouse, will die within a few weeks if deprived of all fluids. We have found no report in the literature of a mammal that can survive more than a few additional weeks by ingesting a three per cent sodium chloride solution. The ability of female mice to tolerate such a regime appears to be outstanding, as the following experiment reveals. Forty-eight young adult female mice were given a stock diet and three per cent sodium chloride solution. The percentage of the animals still living at the end of each month was as follows: first 71, second 60, third 54, fourth 52, fifth 44, sixth 29, seventh 27, eighth 21, ninth 17, tenth 17, eleventh 4, twelfth 0. In the second month four males were introduced. Three were dead within sixty days and the fourth lived approximately twice as long. During this time eight litters were delivered ranging in size from two to nine pups. One additional mouse died during pregnancy. Most of the pups were viable at birth but died in a few days of malnutrition, as lactation was almost nil. Only three pups lived to be weaned and these died within ten days after drinking salt water. Most of

the adult anmals seemed to be in good condition until one or wo weeks before their death, at which time they lost weight and a few developed tremors and ataxia. None showed any evidence of edema.

The effect of diet on liver regeneration in partially hepatectomized rats. EDWARD A. NEWMAN (by invitation) and MORTON I. GROSSMAN. *Dept. of Clinical Science, Univ. of Illinois College of Medicine, Chicago.* Based on the recent works of Denton and Ivy; studies were made to determine whether the enhancement of liver regeneration following partial hepatectomy by feeding liver is due to the high protein content of liver. Four series of experiments were run in the following manner. Partially hepatectomized rats were fed: Group A: 60% casein diet, Group B: coagulated beef heart, Group C: 60% powdered liver diet, Group D: coagulated liver. The rate of regeneration was estimated by sacrificing all the animals eleven days after surgery and calculating the ratio of dried regenerated liver weight to the body weight. Rats fed coagulated hog liver showed a ratio of 0.0115. The next greatest rate of regeneration was seen in rats given 60% powdered liver diet with a ratio of 0.0104. The remaining groups showed a poor regenerative response as seen by the respective order of ratios: coagulated beef heart diet, 0.0078; and 60% casein diet, 0.0082. The average body weight of the rats in all groups was within 5 grams of 300 grams. These results indicate that the enhancing effect of liver is not due to its high protein content.

Electrocardiographic changes on tilting. MICHAEL NEWTON (introduced by H. C. BAZETT). *Department of Physiology, Medical School, University of Pennsylvania, Philadelphia.* Important circulatory adjustments occur on changing from the horizontal to the vertical position. Horwitz (personal communication) noted that a group of patients with possible angina pectoris showed a relative lengthening of electrical systole on tilting to the vertical position and a relative shortening of systole on tilting back to the horizontal. The time relations of these changes were investigated electrocardiographically during studies on the effect of change of environment and other procedures in healthy young men. From Bazett's formula (systole = $K\sqrt{\text{cycle}}$) if K is high, systole is long relative to the cycle, while if K is low systole is short relative to the cycle. On tilting from the horizontal position to 70° from the horizontal the value of K rose to a maximum within 60 seconds; it then decreased to a level which was somewhat above the resting value during the twenty minutes of observation. On tilting back to the horizontal the value of K was greatly decreased; over a five minute period it gradually returned to the resting level. The factors involved in these changes are complex and not easily explained. They are probably chiefly mechanical (venous return) and nervous (vagus and sympathetic influences). The changes observed in the value of K were not significantly affected by changes in environmental temperature, by hemorrhage or by infusion of serum albumin.

Blood volume determination by plasma dye dilution and dilution of red cells tagged with P 32. ROBERT T. NIESET (Introduced by H. S. MAYERSON). *School of Medicine, Tulane University and the Research Division of the Alton Ochsner Medical Foundation.* Independent studies in patients on the rate of absorption and of loss of radioactive phosphorous by red cells in vivo and in vitro and of the loss of phosphorous from the plasma in vivo have been made to check the validity of red cell volume determinations by a simple dilution method using radioactive phosphorous as a tracer. Whole blood samples are used for counting so that no chemical or physical separation of the trace element is required. Discrepancies between whole blood volume calculated from the red cell volumes and that calculated from the plasma volumes as determined by plasma dye (T1824) are analyzed with reference to the ratio of plasma volume to red cell volume. Variations in plasma to cell volume ratios are shown to have opposite effects on the apparent volumes so that the results are not directly comparable. Total blood hematocrits are obtained from the independent measurements of total plasma volume and total red cell volume and are compared with direct hematocrit readings made from peripheral blood. The variation appears to occur in either direction and to be non-specific.

Effect of iodine in hypothyroid rats. WARREN O. NELSON and HELEN E. WHEELER (by invitation). *Department of Anatomy, University of Iowa College of Medicine.* Female rats made hypothyroid by thyroidectomy or administration of thiouracil or thiourea show disturbances of reproductive rhythm, small ovaries and adrenals, and retarded growth. Periods of estrus are very irregular and frequently may be absent for several weeks. When they do occur they are accompanied, as a rule, by ovulation of a few follicles only. It has been observed that the administration of I_2, 6 mg. and KI, 6 mg. in one-tenth cc. water daily will prevent or correct these various departures from normal. A partial effect only was achieved in animals which received 3 mg. I_2 and 6 mg. KI daily. The procedure employed in routinely evaluating the effect of iodine upon hypothyroidism produced by various drugs or by thyroidectomy has involved observation of groups of 12 to 16 animals for 30 days. This serves to establish adequately the hypothyroid state. During the following 30 days one-half of each group receive, in addition, iodine by subcutaneous injection. Estrous cycles usually show improvement within ten days and normal reproductive rhythm is re-established. Ovaries and adrenals are approximately doubled in size and livers, kidneys and spleen show significant weight gains. Iodine-treated animals average approximately 30 grams heavier and 2 cm.

longer at the close of the 30-day period than animals not receiving iodine. The thyroids of rats treated with thiourea or thiouracil plus iodine are smaller than those in rats not receiving iodine. It is supposed that the effects noted are achieved through extra-thyroidal iodination of protein.

Heart temperature and its relation to the T-wave. L. F. Nims, B. Kartin (by invitation), H. M. Chernoff (by invitation) and L. H. Nahum. *Laby. of Physiology, Yale Univ. School of Medicine, New Haven, Conn.* The temperature of the dog's heart was determined by means of thermocouples inserted at several points in the pericardium. The heart temperature averaged about 0.25°C. above rectal temperature, except at the diaphragmatic surface which was about 0.50°C. above rectal temperature. An open chest dropped heart temperature and altered the T-waves in a characteristic manner. On closing the chest, the temperature gradually rose and stabilized after 90 minutes. A pint of hot or cold water placed in the stomach maximally affected the temperature of the diaphragmatic surface. Heat or cold transfer through the anterior chest wall by means of radiant bulb or ice pack maximally altered anterior heart temperature. Various areas of the heart surface were heated and cooled directly by means of a chamber inserted in the pericardial sac in an attempt to quantitate T-wave change in standard and unipolar leads against temperature change. For every 1°C. change in temperature of the left ventricle a 1 mm. change in amplitude of the T-wave was observed, while similar change in temperature of the right ventricle resulted in an average 0.5mm. change in T-wave amplitude, but in the opposite direction.

These experiments suggest that normally occurring temperature differences in various parts of the heart are of sufficient magnitude to account at least in part for the amplitude and direction of the T-wave.

The effect of carbon dioxide on the motility of the small intestine. David W. Northup, J. C. Stickney, and E. J. Van Liere. *West Virginia University Medical School, Morgantown, West Virginia.* Previous reports from this laboratory have shown that anoxia alone has no effect on the motility of the small intestine of the dog (Am. Journ. Physiol. **140:** 119, 1943). In asphyxia, carbon dioxide accumulates in the tissues; therefore experiments were performed in which 6.5–8.0% CO_2 was added to the inspired air to determine the effect of this factor alone—i.e. no anoxia and no asphyxia. In dogs breathing this concentration of CO_2 a charcoal-gum acacia mixture administered by stomach tube traversed an average of 110 cm. of small intestine in 30 min.; in the controls, an average of 165 cm. This reduction in motility is significant at the 4% level.

Influence of anti-organ sera upon the oxygen uptake of the spleen and brain. Wiktor W. Nowinski (introduced by C. M. Pomerat). *Tissue Culture Laboratory and the Psychopathic Hospital, Medical Branch, The University of Texas, Galveston, Texas.* The fact that Reticulo-Endothelial Immune Serum (REIS) in strong concentrations has an inhibitory effect upon the migration of cells in tissue cultures (Pomerat—1946. Quart. of Phi Beta Pi, **42:** 4), suggests the interrelation between this effect and the inhibition of one or several enzyme system. Current investigations were undertaken in order to establish whether anti-organ sera intervene in the oxidative processes of the cell. Oxygen uptake was measured by the usual Warburg technique. The effect of adding anti-organ sera with complement fixation titers ranging from 1:60 to 1:500 were tested at a final dilution of 1:4 and 1:8. Control experiments with corresponding concentrations of normal rabbit serum were carried out smultaneously. In the first series of experiments, anti-rat REIS and rat spleen slices were used. The Q_{O_2} in normal rabbit serum was −14.80, that in REIS, −12.71. As both these figures are very similar and lie in the range of normal oxygen uptake of the spleen, it was concluded that REIS, under these conditions, has no effect upon cellular respiration. Experiments carried out after 3 hours incubation of the rat spleen slices with REIS gave similar results. Likewise, no effect was obtained with concentrations of 1:100, 1:500, and 1:1000, the average Q_{O_2} figures being −10.76, −13.89, and −13.94, respectively. Experiments with chick anti-brain serum showed no inhibition of brain respiration. In these experiments only, brei of chick brain was used and the corresponding average Q_{O_2} was −36.30 with normal rabbit serum as substrate and −33.26 with anti-brain serum. Another series of experiments gave an average Q_{O_2} of −26.78 and −26.03 for normal rabbit serum and anti-brain serum, respectively. Though these figures are somewhat higher than those for the normal respiration of the brain (which possibly might be due to the presence of glucose in the serum), the relative differences between the figures obtained are small enough (10% in the first series) to be ignored.

Inhibition and facilitation of motor activity by the anterior cerebellum. F. E. Nulsen, S. P. W. Black, and C. G. Drake (by invitation). *Laby. of Physiology, Yale Univ. School of Medicine, New Haven, Conn.* Utilization of the inhibitory effect of cerebellar stimulation upon existing motor activity has permitted mapping of the anterior cerebellum as a homunculus representing individual muscles. The cerebral motor cortex or bulbar pyramid are stimulated at regular intervals and the uniform contractions of a single muscle are recorded. Simultaneous stimulation of cerebellar points is then undertaken to establish the location most effective for inhibition of response in that

muscle. Such studies in the cat, dog, monkey, and chimpanzee under dial anesthesia show that the points corresponding to tail, hind-limb, fore-limb, and face musculature have an antero-posterior arrangement in lingula, centralis, culmen, and simplex lobules, respectively. Further localization in the monkey indicates that proximal muscles, as those of the upper arm, are best inhibited by ipsilateral cerebellar points near the mid-line whereas distal muscles, such as the adductor pollicis, are best modified by lateral points in the same lobule. The homunculus for efferent effects thus obtained corresponds to that evolved by Snider and Stowell by observing the localized electrical activity of the cerebellar cortex induced by tactile stimulation.

In each anmal, excepting the cat, increasing the frequency of cerebellar stmulation will result in facilitation rather than inhibition of the existing motor movement. The cerebellar homunculus for such facilitation is identical to that obtained when working with inhibition at low frequencies. The dentate nucleus (probably the dorsomedial portion) is involved in inhibition since its bilateral destruction allows only facilitation on surface stimulation, while its direct stimulation yields pure inhibition at all frequencies. Parallel evidence indicates that the fastigial nuclei transmit facilitatory impulses. Analysis of the effects of additional lesions of the brain stem and cerebellar peduncles, combined with the oscillographic evidence of Snider, Magoun, and McCulloch, suggests that both inhibition and facilitation are ultimately mediated through the bulbar reticular formation.

Thus the same cerebellar point which receives tactile stimuli from a localized somatic area will upon electrical stimulation modify motor activity in the same region of the body. Depending upon the frequency of the stimulus, either inhibition or facilitation can be elicited from the same surface point. Evidence suggests that the pathways for the two effects then diverge to travel through different roof nuclei into the suppressor and facilitatory areas of the reticular formation. It is possible that these results of electrical stimulation may mimic the mechanisms by which the anterior cerebellum modifies the movements of an extremity in response to afferent impulses.

Influence of dibenamine on renal function of the dog. ERIC OGDEN. *Department of Physiology, University of Texas Medical Branch, Galveston, Texas.* Intravenous dibenamine (20 mg./kg.) was given twice to one dog, the doses being separated by a week. The animal immediately appeared nauseated, pale and the jugular veins were empty as in shock. The clearance values were more variable than those immediately before and the urine volumes were diminished. The clearance values observed were as follows: para-aminohippurate before dibenamine 206 (average of 3 days), one week after first dose 183, one week later 18.7 ml./min.; creatinine, 57.5, 45.8, and 7.16 ml./min. In the last experiment the unexpectedly low PAH clearance allowed the plasma concentration to rise to 10.9 mg./100 ml. The clearance therefore may not be a true measure of renal blood flow; calculated as if in the Tm_{PAH} range, the Tm appeared to be 1.19 mg./min. The urine flow was remarkably steady (3.5 to 5.0 ml./min.) and was greater than half the creatinine clearance. This indicates that failure of tubular reabsorption accompanied the failure of PAH excretion. At this time the dog was apparently well but optical records of femoral blood pressure read 174/134 with a pulse rate of 168. Six days later the animal was killed. The kidneys had the gross appearance of tubular degeneration; microscopic findings will be reported. The N.P.N. was 38 mg. per cent post mortem. These findings indicate the need for extreme caution in the use of dibenamine and the matter is being further investigated.

Further studies on the treatment of experimental renal hypertension with paredrine hydrobromide. E. A. OHLER (by invitation) and G. E. WAKERLIN. *Dept. of Physiology, Univ. of Illinois College of Medicine, Chicago.* We have previously reported (Fed. Proc. **6:** 171, 1947) the effect of Paredrine HBr (p-hydroxy-a-methyl phenethylamine) on experimental renal hypertension in seven dogs. Wastl (The Hahnemannian Monthly; Nov., 1942; June, 1944) employed dosages of 0.008 mg. to 80 mg./kg. of Paredrine HBr in renal hypertensive rats and found the more pronounced antihypertensive effect with the lower dosages. Of a total of 12 renal hypertensive dogs treated with 4 mg./kg. of Paredrine HBr orally for periods of from three to seven months, all showed some reduction in blood pressure. One animal gave an excellent response, eight a good reduction, three a fair fall and one a poor response. One neurogenic (debuffered) hypertensive dog showed an excellent reduction. An essential or spontaneous hypertensive dog under treatment at present is exhibiting a good response. Included in the group of eight good responses is one animal which had previously been treated with sodium bromide without effect. One normotensive dog under treatment with 4 mg./kg. of Paredrine orally has failed to exhibit any change in blood pressure. Two other amines, choline and Tuamine (2-aminoheptane), have failed to evidence any antihypertensive effect, each in one dog. Animals under treatment with 4 mg./kg. of Paredrine orally develop a tolerance to the pressor effect of Paredrine as determined by acute intravenous assay on the unanesthetized trained dog. A cross tolerance was exhibited to Tuamine, tyramine, Benzedres (2-amino-1-cyclohexylpropane HCl) and Benzedrine but not to epinephrine,

arternol, ephedrine, renin, hypertensin, or tetra methyl urea.

Renal clearance studies in experimental hypoproteinemic edema. ELIZABETH O'LEARY (by invitation) and SAMUEL A. CORSON, with the technical assistance of OPAL CAIN, CATHERINE ANDERSON and ELINOR FOSTER. *Departments of Physiology, School of Medicine, University of Minnesota and Howard University.* Edema was produced in 5 trained dogs by means of a low-protein diet and periodic massive plasmaphereses. This method permits the production of a fairly standard degree of edema (as measured by the thiocyanate space) within a period of several days. No anesthesia was used. The interval between the control experiments (at normal plasma protein levels) and the experiments under conditions of edema varied from 18 to 32 days. Renal plasma flow and glomerular filtration rates were measured in the postabsorptive state by determining the clearances of sodium para-amino hippurate (PAH) and creatinine respectively. The solution containing these substances was administered intravenously by means of a constant injection pump delivering 1.5 ml./min. A plasma PAH concentration of 2 mg. % or less was maintained by a sustaining dose administered at the rate of 6 mg./M²/min. following a priming dose of 400 mg./M². A plasma creatinine concentration of about 3–4 mg. % was maintained by a sustaining dose of 3 mg./M²/min. following a priming dose of 400 mg./M². The glomerular filtration rate and the renal plasma flow showed a small but consistent decrease under conditions of hypoproteinemic edema. In spite of a decrease in the plasma colloid osmotic pressure (the plasma proteins decreased from an average value of 6.1 g. % to 3.3 g. %), there was no observable increase in the renal plasma filtration fraction. On the contrary, a small but consistent decrease was observed (from a normal average value of 33% to 28% in hypoproteinemia). Incidentally, our average values for glomerular filtration rate (98 ml./M²/min.) and renal blood flow (467) for normal dogs based on creatinine and PAH clearances respectively are in surprisingly close agreement with those reported by Smith (Physiology of the Kidney, Oxford U. Press, 1937, page 262) using inulin (94) and diodrast (433) respectively.

Systolic pressure in the intact, unanesthetized rat. F. OLMSTED (by invitation), A. C. CORCORAN, O. GLASSER (by invitation) and IRVINE H. PAGE. *From the Research Division of the Cleveland Clinic Foundation, Cleveland, Ohio.* An objective means is described for determination of systolic pressure in the intact, unanesthetized rat at normal environmental temperature. Simultaneous graphic records are obtained of the appearance of the pulse in the animal's foot and of the air pressure in an inflatable thigh cuff. The apparatus consists of animal holder, miniature cuff, adjustable platform for the foot, an inelastic cloth band around the foot attached to an electric displacement unit (Statham strain gauge YE-16-250), amplifiers, ink oscillograph, and air pressure recorder. The cloth band is a $\frac{1}{8}$″ wide strip of heavy oiled silk. The ink oscillograph (Brush Development Co.) has a 4″ paper strip on which a record of cuff pressure is obtained from a pen moved by a metallic bellows.

The pulse appears as a rough sinusoidal frequency of 300 to 450 per minute superimposed on a transmitted respiratory movement of 60 to 100 per minute. Inflation of the cuff above systolic pressure obliterates the pulse record. The first appearance of the pulse wave as the cuff pressure is slowly lowered is taken as the systolic pressure point. Pressure is read by calibration of the tracing recorded by the pen connected to the cuff and bellows. Forty six preliminary comparisons were made with optically recorded carotid pressure. Carotid pressure, foot pulse, and cuff pressure were photographed simultaneously. The average deviation was ±5.5 mm. Hg. At arterial pressures ranging from 90 to 150 mm. Hg the average of intra-arterial readings was 117.1, and of foot pulse readings 115.8 mm. Hg.

Methods for extraction of pressor substances from hypertensive blood. NORMAN S. OLSEN (by invitation), HENRY A. SCHROEDER and MELVIN L. GOLDMAN (by invitation). *Departments of Biological Chemistry and Internal Medicine and the Oscar Johnson Institute, Washington University School of Medicine, and Barnes Hospital, St. Louis, Missouri.* Extracts were prepared for rat assay of pressor material from the arterial blood of normotensive and hypertensive patients. One volume of arterial blood was drawn directly into 3 volumes of cold 95 per cent ethanol. Further cold ethanol was added to make a total of 10 volumes. Precipitated proteins were filtered off and 1 ml. of concentrated HCl added to the filtrate for each 200 ml. of original blood. The solute was evaporated to dryness in vacuo below 35°C. The dry residue was extracted with 1 volume of 90 per cent ethanol, filtered and the residue discarded. The filtrate was further concentrated in vacuo to an aqueous residue. This was extracted with petroleum ether which was discarded. The residue was freed of petroleum ether and adjusted to pH 6. This crude extract was found to give pressor effects when injected into rats, but it also contained depressor materials. Using ion exchange resins a separation of the depressor and pressor materials was effected. The latter were found to be present in most extracts of hypertensive blood but not in those from normal subjects. Picrates of the purified material were formed, which on hydrolysis gave pressor effects in rats. From these results it would appear that pressor substances can be extracted from the blood of hypertensive patients.

Effect of pressure breathing and posture upon

the respiratory gas exchange and heart rate. ARTHUR B. OTIS, MITZI SUSKIND (by invitation) and HERMANN RAHN. *Department of Physiology, University of Rochester School of Medicine and Dentistry, Rochester, New York.* Alveolar pCO_2 and pO_2, breathing frequency and volume, and heart rate were recorded at one minute intervals on each of five subjects exposed for 10 minutes to each of the situations listed below. The average % change (without regard to sign) of all measurements is listed for each situation. (Heart rate, breathing frequency and volume, and alveolar pO_2 always increased and pCO_2 decreased.) Although such an averaging together of several diverse measurements is a questionable and inexact procedure, it is done here merely to indicate roughly the relative extent of the changes measured for the various situations.

Supine (Control)	0%
Supine, breathing 20 cm. H_2O (+) pressure	10
Supine, pressure breathing, with pressure vest	5
Supine, pressure breathing, legs bandaged	10
Tilted passively to 70°	13
Tilted, breathing 20 cm. H_2O (+) pressure	37
Tilted, pressure breathing, with pressure vest	32
Tilted, pressure breathing, legs bandaged	31

Tilting and positive pressure breathing act synergistically as shown by the fact that the physiological effects of both acting together are greater than the sum of the effects of each acting alone. Although the protective effects of a pressure vest or of leg bandages against the combined action of pressure breathing and tilting seem small when judged by the physiological data summarized above, they were sufficient to prevent symptoms of nausea and syncope that appeared in 3 of the 5 subjects when no protection was present.

Alterations in fluid and ionic distribution in various patho-physiological conditions. R. R. OVERMAN, C. P. THARP (by invitation) and A. H. TUTTLE (by invitation). *Department of Physiology, University of Tennessee, College of Medicine, Memphis, Tennessee.* Evidence of severe alterations in ionic distribution in febrile disease as indicated by the dilution of SCN in simian and human therapeutic malaria has been previously presented (J. Lab. Clin. Med. **31:** 1170, 1946; Federation Proceedings **6:** 174, 1947). Statistical analyses of data on 70 human patients with therapeutic malaria indicate that alterations in the distribution of SCN, Na, K, and Cl are *not* correlated with (a) progressive anemia, (b) height of fever, (c) length of infection, or (d) parasitemia. There *is* a correlation of high significance between the altered cellular ionic levels and the *number of paroxysms.*

If alteration in body temperature as occurs in the malarial paroxysm is the etiological factor promoting or allowing cellular permeability change, then such changes should be reproducible by artificial fever induction. Measurement of SCN distribution in a dog *during* artificial hyperpyrexia (42°C. rectal for 40 minutes) revealed an 11% reduction in "extracellular" fluid volume. However, if measurement of SCN volume is deferred until the body temperature has returned to normal (8–12 hours later), large increases in the volume of fluid available for SCN dilution occur which are directly correlated with the number of temperature spikes. It has been suspected that the effects of fever on the adrenal cortex might allow such permeability alterations. The results of simultaneous mannitol and SCN volume measurements in unilaterally adrenalectomized dogs tend to bear out this postulate for, while the mannitol volume decreases, SCN volumes increase as they do in the post-pyrexic state.

Influence of age on carbon monoxide desaturation in man. NELLO PACE, ENRIQUE STRAJMAN (by invitation), and ELAINE WALKER (by invitation). *Division of Medical Physics, University of California, Berkeley.* From the work of Roughton and co-workers it is found that, following mild exposure, carbon monoxide is released from the blood of man according to a single exponential function of the form: $CO_t = CO_0 e^{-kt}$. The rate constant, k, may be conveniently expressed as the half time of desaturation of CO from the blood, $CO_{\frac{1}{2}}$, and the latter has been found to vary from approximately three hours while breathing air to one hour while breathing pure oxygen. In the present work, $CO_{\frac{1}{2}}$ was measured in a series of 14 subjects breathing pure oxygen following the attainment of as much as 4.7 volumes per cent CO in the blood. The $CO_{\frac{1}{2}}$ was found to vary from 33.5 minutes to 82.2 minutes, and the age of the subjects ranged from 20 years to 65 years. A raw correlation coefficient of 0.82 between these variables was calculated, and the slope of the line of regression indicates that at the median age of the group, 42 years, there is a 1.0 per cent longer $CO_{\frac{1}{2}}$ for each increased year of age. This value agrees well with that calculated from the data of Berg (Amer. J. Physiol. **149:** 597, 1947) for the influence of age on CO_2 recovery half time following mild exercise. It appears that respiro-circulatory changes occur with increasing age, and are reflected in the rate of gas exchange between blood and ambient air.

The influence of tetraethyl ammonium, hepatectomy and selective destruction of the nervous system on vascular reactivity. IRVINE H. PAGE, R. D. TAYLOR and J. J. REINHARD (by invitation). *From the Research Division of the Cleveland Clinic Foundation, Cleveland, Ohio.* Response to standard doses of adrenalin, nicotine, barium chloride, histamine, angiotonin and renin were studied in normal and anesthetized dogs. Two days after destruction of the spinal cord from C_6 caudad re-

sponsiveness is greatly augmented and it becomes difficult to induce renin tachyphylaxis. Administration of Et_4N now has only a slight augmenting effect. Anterior rhizotomy from C_6 to L_3, or ganglionectomy with subdiaphragmatic vagotomy also increases reactivity, which in such animals is only slightly increased by Et_4N. Removal of the liver in normal animals usually reduces the adrenalin and barium responses; nicotine raises pressure with only occasional lowering while the responses to angiotonin, and especially to renin, are greatly impaired. Even small doses of Et_4N now elicit prolonged pressure fall. The augmentation of pressor substances does not occur. If the kidneys are removed shortly before the liver, the initial response to renin and angiotonin is usually good, but on repeating the dose, the response quickly disappears, not to be restored by Et_4N. Nephrectomy alone increases vascular responsiveness, which is heightened by cord destruction. These preparations are exquisitely sensitive to renin and angiotonin. Et_4N may or may not give further augmentation. Hepatectomy in a dog sensitized by cord destruction decreases the vascular sensitization and the ability of Et_4N to augment. Thus, the nervous system, the liver and kidneys are shown to interplay in determining vascular sensitivity to drugs.

Susceptibility of X-rayed animals to histamine and to adenosine. E. E. PAINTER and M. C. MOORE (by invitation). *Loyola University School of Medicine, Argonne National Laboratory, and University of Illinois College of Medicine, Chicago, Illinois.* A study was made to determine the susceptibility of X-rayed animals to the blood pressure lowering effects of tissue breakdown products, such as histamine and adenosine. The compounds were infused at a constant rate over 4 minute periods. The amounts required to produce a perceptible decrease (5–8 mm. Hg) in carotid blood pressure were determined for the controls and for the X-rayed animals during the first 4 hours after exposure. All animals were anesthetized with pentobarbital before carotid cannulation. Rabbits given a single total body dose of 800 roentgens require only 2–5 μg./kg./min. of histamine base at 1 to 2 hours after radiation to produce a perceptible decrease in blood pressure. At 3 to 4 hours, however, the amount required, 3–8 μg./kg./min., approaches that for the control animals, 10–15 μg./kg./min. With adenosine this change in susceptibility is not so apparent: 70–100 μg./kg./min. for the X-rayed rabbits during the first 4 hours as compared with 90–130 μg./kg./min. for the controls. Preliminary experiments on dogs given a single total body dose of 400 r show a significant change in susceptibility to both compounds. These findings may be correlated with the following changes occurring in the animal body during the early period after exposure to X-radiation and, therefore, may be related to radiation sickness: 1) liberation of tissue breakdown products, 2) alteration in the detoxifying powers for such substances, and 3) alteration in the compensatory mechanisms for reducing the effects of such substances.

Certain physiological reactions to intense sound fields. H. O. PARRACK and D. H. ELDREDGE, JR. (introduced by H. M. SWEENEY). *Aero Medical Laby., Air Materiel Command, Wright Field, Dayton, Ohio.* The response of the ear and other sensory mechanisms to sound fields at a level of 140 to 160 db above 0.0002 dynes per square centimeter will be described. Exposures have been made, at this level, to essentially single frequencies in the range from 5 Kc to 35 Kc. Pain and thermal sensations, hearing loss with suitable exposure frequency, and other sensory reactions are induced. Physiological reactions to intense broad spectrum sound fields will also be described. These fields are of the order of 140 db above the reference level 0.0002 dynes per square centimeter and consist of a continuous sound spectrum from approximately 50 cps to 50,000 cps. Hearing loss occurs together with disturbance of the posture maintaining mechanisms.

Secretion and excretion of carbohydrate-active adrenal compounds (oxysteroids). K. E. PASCHKIS, A. CANTAROW, A. A. WALKLING (by invitation), W. H. PEARLMAN, A. E. RAKOFF (by invitation) and D. BOYLE (by invitation). *Jefferson Medical College, Philadelphia.* Study of metabolism of adrenal steroids has so far been mostly concerned with the urinary excretion of androgens and related compounds (19 carbon atom compounds) in humans. Experiments are presented investigating the secretion and excretion of carbohydrate-active compounds (oxysteroids) in dogs and rats using the mouse liver glycogen assay method. No activity was found in peripheral blood of dogs. Blood from the adrenal vein obtained through a London-cannula showed high activity. This confirms data in the literature obtained under less physiological conditions of acute experiments. Active material is found in the urine of dogs but not of rats. After injection of adrenal cortical extract active material is excreted in bile and urine of dogs.

Influence of temperature on radio-sensitivity in the frog (Rana pipiens). HARVEY M. PATT, MARGUERITE N. SWIFT and ELLA B. TYREE (introduced by AUSTIN M. BRUES). *Biology Division, Argonne National Laboratory, Chicago, Illinois.* There is considerable evidence which suggests that radio-sensitivity of tissue is dependent upon certain aspects of its activity (growth rate, temperature, blood flow). Since temperature effects on biological processes are most evident in poikilothermic animals, we have used the frog to study the influence of temperature on survival after

X-irradiation. In the preliminary experiments reported here animals were given total body X-radiation at 23°C. and were kept at different temperatures after irradiation. Survival of experimental animals was corrected for the small mortality observed among non-irradiated controls.

Dose	Temp.	No. frogs	% survival—weeks after x-irradiation						
			1	2	3	4	5	7	18
r	°C.								
3000	22	59	80	63	27	9	7		
3000	12	10	90	80	70	70	70		
3000	5	40	100	100	100	100	100		
6000	23	12	100	0					
6000	5	12	100	100	100	100	100	100	100
9000	22	12	100	0					
9000	5	12	100	100	100	100	75	0	

The prolonged survival with low temperature treatment is clearly evident. Some 80–90 per cent of animals kept at 5°C. for periods of three to four months after irradiation have survived a dose which is lethal to animals at 22°C. within three to six weeks. Experiments are in progress to determine whether the altered radio-sensitivity is accomplished merely by a decrease in the rate at which degenerative changes occur or by a more rapid and complete recovery from the effects of irradiation.

A negative form of the oculo-gravic illusion. J. L. PATTERSON, JR. and ASHTON GRAYBIEL (introduced by JAMES V. WARREN). *Naval School of Aviation Medicine and Research, Pensacola and Department of Physiology, Emory University Medical School.* The otolith organs of the non-auditory labyrinth are believed to be the proprioceptors involved in the sense of static position. If a subject remains in fixed position during acceleration on the human centrifuge, the direction of physical force will be changed in relation to him. This force is the resultant of the force due to gravity, centrifugal force and tangential force. In the dark, this situation produces a sensation of bodily tilt in space. A small illuminated visual target, fixed in relationship to the subject, will appear to undergo corresponding displacement (oculo-gravic illusion). The present studies were conducted on the centrifuge with the subject fixed in the sitting position in the dark and continuously observing a free-swinging luminous pendulum. This pendulum indicated at any given moment the direction of resultant physical force. Twenty-one experiments on four subjects were done, in each of which the subject was taken from rest to a maximum of 12 RPM and back to a position of rest. At 12 RPM the resultant force was 1.3 G, directed 39° 49′ from the true vertical.

Throughout the experiments the direction of the pendulum appeared to remain vertical or very nearly so. This was true even when the pendulum was directed approximately 40° from the true vertical. It is concluded that, in the absence of visual cues and within the limits of the forces produced in these experiments, the apparent vertical is represented by the direction of resultant physical force.

The effect of oöphorectomized dogs' urine extract on gastric secretion. T. L. PATTERSON, J. KAULBERSZ, D. J. SANDWEISS and H. C. SALTZSTEIN (by invitation). *Depts. of Physiology and Surgery, Wayne Univ. College of Medicine and Harper Hospital, Detroit, Michigan.* Urogastrone from normal dogs inhibits gastric secretion in the majority of experiments while a similar extract from hypophysectomized dogs stimulates gastric secretion. In a limited number of studies it was also found that an oöphorectomized urine extract inhibited gastric secretion very little if at all. To check the latter findings 41 additional experiments were performed with oöphorectomized urine extracts on 5 pouch and 4 fistula dogs. The method of preparation, the yield of the purified extract and the dosage applied were similar to those previously reported. The average output of free HCl after injection of oöphorectomized extracts differed only 10.8 per cent from the averages of the control experiments; when administered in the first period an average stimulation of 9 per cent resulted; when given in the second period an increase of 13 per cent was found as compared to the controls. These differences were so close to the variations of the effect of histamine alone at different times that they can not be considered as significant. One might therefore assume that the urine extract from oöphorectomized dogs is producing neither inhibition nor stimulation of the gastric juice, following histamine injections. It appears that the ovaries may play a rôle in urogastrone production. However, it is possible that the effect of oöphorectomized urine extract is due to the dependence of the ovaries from the regulatory mechanism of the pituitary gland, since the latter has been found to exert some influence on the formation or excretion of urogastrone.

Instability of motor points and sensory points in the human cerebral cortex. By WILDER PENFIELD and KEASLEY WELCH (by invitation). *McGill University, Montreal, Quebec, Canada.* Sherrington and his pupils analyzed mechanisms by electrical stimulation of the cerebral cortex of anthropoid apes. The human cerebral cortex reacts in a similar manner to such stimulation. There is instability of cortical points, i.e., variation in the response of the same point. In man this may be

shown to apply to sensory points as well as motor. By means of successive stimulations the threshold may be lowered and a given motor response displaced to a distance. This results in deviation and reversal of responses. The limits of sensory and motor stimulable cortex will be discussed.

The effects of changing the temperature of the normal and denervated nictitating membrane. JOHN F. PERKINS, JR. and M. C. LI (introduced by E. M. LANDIS). *Department of Physiology, Harvard Medical School, Boston, Mass.* The temperature of the nictitating membranes of cats was changed by means of glass eyes with water circulating through them substituted for the normal eyes. Heights of contraction were determined at water temperatures ranging from 10 to 44 degrees centigrade in 5 degree steps. Upon stimulating the cervical sympathetic, maximal shortening, taken as 100 per cent, occurred at 30 degrees, dropping to an average of 57 per cent at 44 degrees and to 85 at 10. Without electrical stimulation cooling to 10 degrees produced 61 per cent shortening, warming to 44 degrees gave minimal, i.e. zero per cent shortening. Evidence from epinephrine dose-response curves suggests destruction of sympathin at the higher temperatures rather than decreased contractility. Effects of cooling were studied by suddenly, or gradually, lowering the temperature of the water from 37 degrees by varying amounts. The denervated nictitating membrane contracted in jerky, stepwise fashion when cooled, in contrast to the smooth contraction of the normal. When heights of contraction were plotted against degrees of sudden cooling the shape of resulting curves was different from that of dose-response curves for epinephrine, suggesting separate response mechanisms. When cooling and epinephrine were tested simultaneously the response equalled the sum of the two effects when tested separately, no true synergism existing. In two of four experiments the denervated nictitating membrane showed true sensitization to cold, contracting in response to smaller drops in temperature than the normal and showing greater contractions with a given drop in temperature for all but maximal contractions.

Effect of insulin upon the in vitro glucose utilization and glycogenesis of the diaphragm of normal and pituitarectomized rats. Adaptation of this technique as an assay for serum content of insulin and anti-insulin substances. M. PERLMUTTER (introduced by G. W. THORN) and R. O. GREEP. *Department of Medicine, Harvard Medical School, the Medical Clinic, Peter Bent Brigham Hospital, and the Harvard School of Dental Medicine, Boston, Mass.* Gemmill has shown that insulin increases the glucose utilization and glycogen synthesis in the isolated rat diaphragm. Using the same in vitro technique, studies were made of the insulin effect on the diaphragm of normal and pituitarectomized rats. The experiments were done on the fourth and fifth day after hypophysectomy. The insulin effect on the glucose utilization was not decreased in the hypophysectomized tissue. By modifying this technique, an assay for insulin content of human serum has been devised. Glycogen synthesis in a mixture of normal serum and 0.5 per cent glucose in Krebs phosphate medium was 0.32 ± 0.014 (S.E.M.) mg. per cent. Further work is being done, comparing the effect of sera from normal and hypoglycemic humans to determine the value of this test in the diagnosis of hyperinsulinism. The addition of 0.01 units of insulin per ml. of serum in vitro or the injection of 20 units of crystalline insulin intravenously into the human significantly increases the glycogenetic effect of the serum. By the use of this method, a search for anti-insulin factors in the sera of insulin resistant diabetics and patients with Cushing's syndrome is being made.

Effect of graded degrees of muscular tension on human heart rate. JOHN E. PETERS (by invitation) and W. HORSLEY GANTT. *Pavlovian Laboratory of the Phipps Psychiatric Clinic, Johns Hopkins Hospital.* These experiments were designed to show the changes in heartrate with accurately measured muscular tensions in humans. Previous studies from this laboratory revealed that even slight emotional states—those produced by a conditional stimulus for a small amount of food (2 gms.) or for faradic shock—are accompanied by cardiac acceleration. How much of this results from increased muscular tension, how much from central nervous excitation independent of peripheral tension? In 9 normal subjects exerting a) maximal tension with left hand on a dynamometer 5 seconds, b) maximal tension 30 seconds, c) one-sixth maximal tension for 2 minutes, there was: 1) great individual variation [56–95 control; 68–116 with a); 90–136 with b); 66–117 with c)]; 2) accelerated heartrate with a) and b) [from 82 control to 97 test with a) and from 85 control to 112 test with b)]. With one-sixth maximal tension no change in heartrate occurs during the first 30 seconds; a moderate increase during the last 90 seconds coincided with fatigue. The muscular tension produced with one-sixth of maximal is nearer the muscular tension during a conditional reflex—considerably less than our maximal tension. Respiratory changes were much less than those accompanying a conditional reflex having a fairly strong underlying motivation.

From these and other experiments we conclude that an increased heartrate occurring with a salivary or even with a motor conditional reflex is much more a result of central nervous excitation than of any accompanying peripheral muscular tension.

Beneficial effects of calcium chloride in fluoride

poisoning. J. H. PETERS (by invitation), L. GREENMAN (by invitation), and T. S. DANOWSKI. *From the Department of Research Medicine and the Renziehausen Foundation, University of Pittsburgh, School of Medicine.* 1.0 cc. of a 5 per cent solution of sodium fluoride in water was administered by gavage to immature female rats weighing 60 to 115 grams. Without treatment death occurred in 24 hours or less in 75 to 95 per cent of the animals in a series of experiments. The introduction into the stomach of 3.0 cc. of a 4 per cent solution of calcium chloride in water just prior to, or within 5 minutes after, the sodium fluoride reduced the mortality to less than 10 per cent. A delay of 10 minutes in the administration of the calcium chloride to fluoride-poisoned rats reduced significantly the efficacy of this treatment. Tetany, a frequent though not invariable manifestation in the rats given sodium fluoride, disappeared with calcium chloride therapy in all but the moribund animals. The beneficial effects of calcium chloride in fluoride poisoning are related, in all probability, to the removal of the fluoride ion from solution as the highly insoluble salt, calcium fluoride. At the same time the hypocalcemia and tetany which develop in fluoride poisoning are corrected.

Direct blood pressure recording in man. L. H. PETERSON and G. C. RISMAN (introduced by H. C. BAZETT). *Department of Physiology, School of Medicine, University of Pennsylvania, Philadelphia, Pa.* An innocuous, mobile, practical blood pressure recording device has been developed by combining with a Lilly capacitance manometer the catheterization technique described a year ago and an ink writer—d.c. amplifier couple. The response is critically damped and linear from d.c. to one hundred cycles up to 300 mm. Hg. Intra-arterial pressure pulses have been recorded from the human continuously under a wide variety of circumstances including surgical and other clinical procedures. To date over seventy-five patients have been followed through anesthesia and surgery. Congenital heart defects and normal humans under physiological stresses have also been studied. The technique does not interfere with the patient's comfort or the surgeon's convenience.

Correct manometric records have been obtained continuously for up to ten hours without clotting or flushing. One individual can accomplish the entire procedure. Calibrations and base line determinations can be made at will. Under such circumstances it is easy to see the wave shape and correct pressures of each beat as it occurs. Deductions can be made, when using each individual as his own control, whether changes in pressure or pulse shape are due primarily to imbalance of cardiac output, peripheral resistance or arterial distention. Such immediate knowledge greatly aids correlation of causes and effects. Surgery, treatment or experimental procedure can be modified according to the effects observed and valuable time may be saved both in surgery and in repetition of experiments. Data are presented to demonstrate specific examples of such values.

The effect of adrenalectomy or evisceration on cyclopropane induced cardiac arrhythmias in cats. FRANK L. PETTINGA (by invitation) and J. W. STUTZMAN. *Department of Pharmacology, Boston University School of Medicine.* After induction with a cyclopropane-oxygen mixture unanesthetized cats were intubated and maintained in deep surgical anesthesia by rebreathing a mixture of 28 per cent cyclopropane in oxygen from a 100 liter reservoir. The beam of the electrocardiograph (Lead II) was under constant observation. Records were taken every two minutes and when a change of rhythm was observed. Control experiments consisted of recording the cardiac response during 30 minutes of cyclopropane anesthesia. On a subsequent day the animals were again placed on the anesthetic mixture. As soon as a record of cardiac arrhythmias was obtained bilateral adrenalectomy or abdominal evisceration was performed. The finding of Allen et al. (Anesthesiology **3:** 530, 1942) was confirmed that without exogenous epinephrine cats develop cardiac arrhythmias during cyclopropane anesthesia. The arrhythmias consist of ventricular premature contractions, bigeminal rhythm, and ventricular tachycardia. Bilateral adrenalectomy caused the preexisting ventricular tachycardia to revert to S-A rhythm within two minutes in 5 of 6 animals. In the sixth a normal rhythm followed evisceration. Removal of the spleen, gastrointestinal tract and accompanying mesentery from cardia to rectum abolished the ventricular arrhythmias in 5 of 7 cats. Visceral arteries were ligated within 2 cm. of the aorta. Adrenalectomy restored a normal rhythm in one of the remaining two, and splanchnicectomy in the other.

Variations with age in neutral steroid excretion of men. GREGORY PINCUS, LOUISE ROMANOFF (by invitation) and JAMES CARLO (by invitation). *The Laboratories of the Worcester Foundation for Experimental Biology, Shrewsbury, Massachusetts.* Data have been collected on over one hundred normal, healthy men of various ages on the urinary 17-ketosteroid excretion. For basal (sleep) urines there is no significant difference in the mean excretion values between the 3rd, 4th and 5th decades of life. A decline in mean excretion values is apparent at the 6th decade which continues on into the 9th decade. The diurnal rhythm of excretion of 17-ketosteroids is scarcely altered in the older age-groups. Data on the urinary excretion of neutral reducing lipids reveal no change in mean basal values through the 8th decade. A more irregular diurnal rhythm of excretion appears in the older

men. Since both of these measures reflect adrenocortical secretion it is concluded that those physiological activities influenced by the neutral reducing lipid precursors are constantly maintained in healthy men whereas the physiological activities influenced by 17-ketosteroid precursors diminish in the later decades of life.

The effect of vagotomy on the secretion of pancreatic juice after the ingestion of various foodstuffs. (Preliminary Report.) I. J. PINCUS (by invitation), J. E. THOMAS and P. O. LACHMAN (by invitation). *Jefferson Medical College, Philadelphia, Pennsylvania.* In view of the current interest in the effect of vagotomy on gastro-intestinal function, it was thought timely to present this report. Feeding experiments were performed on three dogs to determine the normal pancreatic response to various foodstuffs. Milk, meat, bread and olive oil were fed in quantities which represented approximately equal amounts of total solids. The volume and specific gravity of the pancreatic juice was determined and the nitrogen out put calculated. These experiments were then repeated in one animal after a successful transthoracic vagotomy. Following vagotomy, the volume of pancreatic juice was reduced to a significant degree. The average hourly secretion for the five hours after feeding 250 ml. of milk dropped from 18.6 to 7.8 ml.; when 55 grams of meat was fed, it decreased from 23.4 to 9.1; whereas 35 ml. of olive oil produced an hourly secretion of 17.9 ml. before vagotomy, it caused an hourly flow of only 11.1 ml. after vagotomy; 50 grams of bread, which had caused an hourly flow of 23.1 ml., produced only 8.2 ml. after vagotomy. The specific gravity of the pancreatic juice was reduced, and the total nitrogen output was markedly diminished.

It would seem that after vagotomy, the secretion of pancreatic juice in response to food is decreased in amount; the enzyme concentration which parallels the specific gravity is low, and the nitrogen output is markedly reduced. It may be that some of the complications observed when vagotomy is used as a therapeutic measure are produced by this effect on pancreatic function.

The reabsorption and excretion of bicarbonate in normal man. By R. F. PITTS, J. L. AYER and W. A. SCHIESS. *Dept. of Physiology, Syracuse University College of Medicine, Syracuse, N. Y.* In a series of 14 experiments on 3 normal male subjects in whom plasma bicarbonate was elevated to 38 mM. per liter by infusing sodium bicarbonate, from 2.6 to 3.0 mM. were reabsorbed per 100 ml. of glomerular filtrate. The excess appearing in the filtrate was excreted. At plasma concentrations of 13 to 23 mM. per liter (following ingestion of ammonium chloride) reabsorption of bicarbonate was 99.9+ per cent complete. Thus the normal renal threshold for bicarbonate in our experiments varied between 24 and 26 mM. per liter. It is suggested that the mechanism for the reabsorption of bicarbonate in the distal tubular segment is the same as that which converts buffer salts into free titratable acids by the exchange of hydrogen ions for fixed base. If this same exchange were effected for base bound by bicarbonate, one would predict first, that the partial pressure of carbon dioxide in the urine (i.e. concentration of free carbonic acid) should increase in proportion to the rate of distal tubular reabsorption of bicarbonate; and second, that the rate of excretion of titratable buffer acid should be inversely related to the rate of distal tubular reabsorption of bicarbonate; i.e. carbonic acid should substitute for buffer acid. The experimental data are in accord with both predictions. It is probable that the proximal tubular mechanism reabsorbs bicarbonate isotonically, i.e. in proportion to water. If true, the volume of fluid reabsorbed in the proximal segment must be inversely related to plasma bicarbonate concentration.

Blood and urine levels of theophylline after intravenous injection. A. J. PLUMMER (introduced by GEORGE L. MAISON). *Department of Pharmacology, Boston University School of Medicine.* A method for the determination of theophylline has been developed and will be reported in detail elsewhere. Theophylline is extracted from a trichloracetic acid blood filtrate, precipitated as a copper compound in methyl alcohol solution and determined by titration of the iodine liberated from a soluble iodide by the copper compound. The method is sensitive to 0.1 mg. per 100 cc. blood. It has been applied to blood and urine. Theophylline was determined in the blood of five dogs after the intravenous injection of 10 mg. per kg. of theophylline with ethylene diamine at intervals of five, thirty, sixty, and ninety minutes after the injection. The levels, expressed in mg. theophylline per 100 cc. of blood are as follows: at 5 minutes 0.76 to 1.10; at 30 minutes 0.43 to 0.58; at 60 minutes 0.16 to 0.30; at 90 minutes 0.10 to 0. Theophylline was detected in only one animal's blood after 90 minutes. The theophylline in the urine was determined in catheterized specimens taken at thirty, sixty and ninety minutes after the intravenous injection. During the first thirty minute period the average excretion was 0.062 mg. per kg., during the second thirty minute period 0.046 mg. per kg. and during the third period 0.036 mg. per kg. During the 90 minute period 1.8% of the injected drug was excreted.

Venous pressure in the human leg during exercise and in various positions. ALBERT A. POLLACK and EARL H. WOOD. *Section on Physiology, Mayo Clinic, Mayo Foundation, Rochester, Minnesota.* Direct and continuous measurement of the pressure in the great saphenous vein at the ankle in eleven normal subjects in the supine, sitting and

standing positions and while the subjects were walking on a treadmill was made by means of a strain-gauge manometer connected to a polythene tube which was inserted in the vein through a no. 17 needle. The average pressure in the supine position was 11.6 (7–15.9) mm. of mercury. In the sitting position, the average pressure was 55.9 (45–67.5) mm. of mercury and in the resting standing position was 86.8 (78.5–92.6) mm. Resting venous pressure at the ankle in the various positions studied was sufficient to support a column of blood up to the third thoracic interspace at the sternum. Taking a single step produced an average initial rise of 10 (− 3 to 28) mm. of pressure even before the heel was lifted from the floor. An average fall of 51.8 (28–80.5) mm. of mercury occurred while the foot was off the floor. As the heel returned to the floor, there was a slight rise in pressure, followed by another fall to the previous low level as the heel settled on the floor (and the body weight was redistributed). As the subject stood quietly after taking the step, the pressure returned to the control level over a period of 22.9 (11–37) seconds. While the subjects were walking on a treadmill at 1.7, 2.6 and 3.3 miles per hour, the pressure fell in all subjects in the first four to twelve steps to an average mean stable value of 22.3 (11–30.6), 24.3 (8.5–38) and 23.5 (10–43.2) mm. of mercury respectively and returned to the control level in 31.2 (8–57) seconds after the walk. Inclining the treadmill to 20 degrees made no difference in the fall of pressure during the walk.

The toxicity of various stimulants of nervous tissue as revealed by tissue culture technique. J. PAINTER and C. M. POMERAT., *Tissue Culture Laboratory of the Department of Anatomy and the Psychopathic Hospital, Medical Branch, The University of Texas, Galveston, Texas.* A technique has been devised to observe the outgrowth of nerve fibers and glial elements from explants of spinal cords of 9-day chick embryos in hanging drop preparations. The toxicity of various barbiturates has been studied with this method (Pro. Soc. Exper. Biol. and Med., **63**: 322, 1946).

Total inhibition of fiber outgrowth resulted from the inclusion of 500 γ/ml. of benzedrine sulfate while a few short fibers were seen in medium containing 250 γ and cultures with 150 γ/ml. of this drug resembled the controls. Metrazol was studied with the use of a 10% solution ordinarily employed for clinical purposes (Bilhuber-Knoll Corporation). Total inhibition was obtained with a 1:8 dilution of 1:10 (10,000 γ). Picrotoxin proved remarkably nontoxic. Outgrowth from explants in a medium containing 3.75 mgs./ml. appeared uninjured. The migration of fibers was not damaged in medium containing 1000 γ/ml. of camphor tetrazol but the addition of 1250 γ/ml. proved totally inhibitory. Crystalline and protamine-zinc insulin at 10 γ/ml. permitted outgrowth of fibers resembling corresponding untreated control cultures for the first 24 hours, but by 48 hours it was evident that these elements were arrested. Stained preparations revealed only fibers of relatively large diameter, the dense network of fine fibers being almost completely eliminated.

Uropepsin excretion in man. C. J. PODORE (by invitation), R. H. BROH-KAHN (by invitation) and I. ARTHUR MIRSKY. *May Institute for Medical Research of the Jewish Hospital, Cincinnati, Ohio.* Studies have been undertaken to determine some of the various factors that influence the rate of uropepsin excretion in man. It was first essential to demonstrate that the activity of uropepsin in the urine specimens as they are tested in the laboratory reflects the true concentration of this enzyme as it is excreted in the urine. The stability of uropepsin was confirmed and no uropepsin inhibitors were found in urine. The urine from numerous normal subjects was collected over a 24 hour period for several weeks without interruption. In many cases the urine excreted during the hours of activity was collected separately from that excreted during sleep. No correlation could be observed between the excretion of uropepsin and the acidity of the urine or the rate of urine excretion. Uropepsin was excreted at a fairly constant rate over a 24 hour period and was not influenced by urine volume so that it was the total amount of uropepsin appearing n the urine rather than its concentration that characterized its excretion. Ordinary fluctuations in daily diet failed to produce any marked variation in excretion. Nocturnal excretion in general was somewhat lower than excretion during the day but much overlapping was observed. Each subject excreted uropepsin in a fairly characteristic pattern with but fairly small daily fluctuations. Children were characterized by a lower uropepsin excretion than were adults. The excretion of patients with gastric lessons was compared with those of normal subjects. The great majority of patients with benign peptic ulcer fell into a group that was characterized by a higher daily mean uropepsin excretion than that of the normal. Fluctuations about this mean were greater in the ulcer group than in the normals. Patients with malignant ulcers, as verified by pathological examination, were characterized by a somewhat lower uropepsin excretion than the normal subjects.

Variation of total circulating hemoglobin and reticulocyte count with season and following hemorrhage. R. L. POST (by invitation) and C. R. SPEALMAN. *University of Pennsylvania, Medical School (Physiol. Dept.).* Total circulating hemoglobin was determined by the carbon monoxide method using a filter photometer on four men at about two-week intervals from June 1946 through August 1947. Reticulocytes were counted at similar

intervals from December 1946 through August 1947. The mean monthly outdoor temperature ranged from 30°F to 70°F in this period. During the summer of 1946 increases in total circulating hemoglobin from 6 to 11% were observed but in 1947 no significant increase was found. Similarly no increase in reticulocyte count was found in 1947. These results need not be inconsistent with reports in the literature of a reticulocytosis in the spring and an increase in total hemoglobin in hot weather because these subjects spent most of their time indoors. To determine the reticulocytosis produced by a sudden small deficit of hemoglobin four other subjects were bled 10% of their blood volumes and total hemoglobins and reticulocyte counts were determined almost every day for more than two weeks. The hemoglobin deficit was approximately one-half restored in two weeks. The reticulocyte count rose almost linearly from a normal of 7 per 1000 rbc to a peak of 18 per 1000 rbc in 14 days.

The reticulocyte count cannot be taken as directly proportional to hemoglobin production under these circumstances. On the basis of our data it would appear that either the maturation time of the reticulocytes must increase or that a higher proportion of red blood cells leaving the marrow must be in the reticulocyte stage following such a hemorrhage.

Action of narcotics on synapses compared to action on axons in sympathetic ganglia. J. M. POSTERNAK (by invitation) and M. G. LARRABEE. *Johnson Foundation, University of Pennsylvania.* The action of narcotics on synaptic functions were compared with effects on axonal conduction by recording impulses transmitted through perfused stellate ganglia of cats. The postsynaptic action potential in the inferior cardiac nerve following preganglionic nerve stimulation measured the number of cells responding to synaptic excitation; the action potential of the cervical sympathetic trunk measured the number of type B fibers conducting impulses through the stellate ganglion without synapse. Chloroform and ethyl ether exhibited preferential action on the synaptic pathway, which was depressed by concentrations about ½ those which equally depressed conduction over B-fibers. Both narcotics caused detectable effects at concentrations equalling those in blood for human surgical anesthesia. By contrast, ethyl alcohol (previously reported) and methyl alcohol had no preferential action on synapses. Actually axonal conduction was depressed in concentrations about ½ the concentrations equally depressing the postsynaptic response. Higher aliphatic alcohols exhibited a pronounced differential action, octyl alcohol depressing the synaptic pathway in ½ the concentration required for equal effect on B-fibers. This preferential action developed progressively in the aliphatic series (C_1 to C_6 and C_8), the ratio of required concentrations being a linear logarithmic function of the number of carbon atoms.

For a given effect on the synaptic pathway, thermodynamic activities were nearly equal for ethyl ether, chloroform and the alcohols, while molar concentrations varied nearly 6000 fold. For effect on B-fibers, the thermodynamic activities increased ten-fold with number of C atoms in the alcohols, the activities for chloroform and ethyl ether being intermediate.

A simple calorimeter for simultaneous determination of heat production and heat loss in laboratory animals. LAWRENCE R. PROUTY (by invitation), Martha J. Barrett (by invitation) and JAMES D. HARDY. *From the New York Hospital and the Department of Physiology, Cornell University Medical College, New York, New York.* The calorimetric technique developed by Day and Hardy for premature infants has been improved to give a rapid method of simultaneous measurement of respiratory metabolism and heat loss in animals. Expensive equipment and involved procedures heretofore required for direct heat determinations have been largely eliminated. Heat production and heat loss in calories per hour can be measured in single periods with an accuracy of ± 2%. A single observer using half hour periods can operate the entire apparatus. The calorimeter consists of two concentric copper cylinders separated by a 10 mm. air layer. Air is circulated through the inner cylinder (the calorimeter proper). Thermocouples are placed at intervals over the surface of each cylinder and in the ingoing and outgoing air stream apertures. Within and insulated from the inner cylinder are a wire mesh floor, skin, muscle and rectal thermocouples and electrocardiograph leads. Activity of the animal is recorded from a pneumograph attached to the suspended calorimeter and from electrocardiograph changes. Direct calorimetry is based on the measurement of thermal gradient between the two copper cylinders, the outer of which is cooled to room temperature by an electric fan. Correction is made for vaporization and calorimeter storage under non-equilibrium conditions. The calorimeter is sufficiently versatile to be used with either closed or open circuit method. We found the open circuit, gravimetric method of Haldane sufficiently accurate for general use. Where small quantities of oxygen and carbon dioxide are present the gasometric method of Barker is more applicable.

Man's respiratory response during acclimatization to high altitude. HERMANN RAHN and ARTHUR B. OTIS. *Department of Physiology, University of Rochester School of Medicine and Dentistry, Rochester, New York.* The changes in alveolar air composition during acute exposures to high altitude are compared to those recorded in the literature for

acclimatized individuals. A diagram is presented which allows one to visualize the simultaneous alveolar oxygen and carbon-dioxide pressures as well as the alveolar ventilation at any altitude and to predict the magnitude of the changes in these three quantities during the process of acclimatization. The prediction for changes in alveolar air were verified by exposure of three subjects to 9500 ft. for several weeks.

With the assumption that the blood pH returns to normal when acclimatization is complete, one can estimate that the alkali reserve at this altitude is lowered approximately 20%. If the pH has a regulatory action upon the ventilation then the reduced alkali reserve should make the respiratory system more sensitive to equal increments in CO_2. This is indicated by (1) a greater ventilation response to 22 and 34 mm. pCO_2 in the inspired air after acclimatization, (2) a smaller increment in alveolar pCO_2 necessary to achieve the breaking point after breath holding when pure oxygen was breathed at altitude and, (3) by reduction in breath holding time when breathing pure oxygen at altitude.

Dynamical and electrical features of human isolated voluntary muscle in isometric and isotonic contraction. H. J. RALSTON, J. R. CLOSE (by invitation), V. T. INMAN (by invitation) and B. FEINSTEIN (by invitation). *Dept. of Physiology, College of Physicians and Surgeons, San Francisco, and the University of California School of Medicine and College of Engineering, San Francisco and Berkeley.* Mechanical and electrical responses of essentially isolated voluntary muscle have been studied in amputees having cineplastic muscle tunnels. Stimulation of the muscles was in all cases voluntary. Isometric length-tension diagrams reveal the same basic features as those shown by frog muscle. Since force developed varies with length of muscle, no correspondence between mechanogram and electromyogram exists, except at a given length of muscle. Isometric contraction times are much greater than those in artificially stimulated muscle-nerve preparations, in some cases exceeding one second. The slow development of both force and electrical activity in voluntary contraction indicates that voluntary response is at first fractional. At a given length of muscle, forces developed in isotonic contraction are only a few per cent of those developed in isometric contraction. Velocities of free contraction as great as six times the resting muscle length per second have been observed.

The effect of eserine on the absolutely and relatively refractory period of turtle heart strips. ROBERT W. RAMSEY, GEORGE FISCHER (by invitation) and MARIE-LOUISE FLINKER (by invitation). *Department of Physiology and Pharmacology, Medical College of Virginia, Richmond, Virginia.* Turtle heart strips (ventricle) were equilibrated overnight in Ringer's solution, and on the following day stimulated electrically at a constant rate of 6 per minute. The temperature ranged from 25 to 30°C. Short condenser shocks (cr = 5 milliseconds) were applied at various intervals between two successive ten second responses and the voltage required to cause an additional response recorded. Adequacy of response to the interpolated shock was judged by the appearance of a second action potential. When the voltages of the interpolated shocks are plotted against time measured from the preceding shock, the resultant curve maps out both the absolutely and the relatively refractory periods. Such curves were obtained first in Ring and then after application of the Ringer-eserine solution. In a few experiments the curves were redetermined after washout of the drug by Ringer's solution. The concentrations of the eserine solutions used (either as salicylate or sulfate) ranged from 1/100,000 to 1/10,000. In the majority of instances eserine slowed the rate of recovery of the threshold to its initial value and thus prolonged the relatively refractory period. There was no experiment (out of 18) in which the drug shortened the relatively refractory period. The drug did not significantly affect either the absolutely refractory period or the initial threshold.

Ventricular isometric relaxation phase as measured on the electrokymogram. EDWARD F. RANDAK (by invitation), BERT R. BOONE (by invitation), GEORGE F. ELLINGER (by invitation), and M. J. OPPENHEIMER. *Dept. of Physiology, Temple Univ. School of Medicine and Heart Disease Control Section, U. S. Public Health Service, Philadelphia, Penna.* The development of the electrokymograph (Am. Jr. Roentgen. and Rad. Therapy **57**: 409, 1947), has provided a new method for studying the pulsations of the heart and great vessels on the intact human subject. Clearly recorded serrations following the ventricular ejection limb appear to provide an easy means of measuring the duration of the isometric relaxation phase. Little importance has been given this phase in past investigations. Wiggers and Burstein, utilizing optical venous and arterial tracings on man, have provided some figures with which those of our direct method are in close agreement.

	Range	Average
	seconds	*seconds*
Wiggers and Burstein	0.037–0.136	0.076
Randak, et al.		
96 normals	0.060–0.160	0.113
22 abnormals	0.100–0.220	0.178

There is little difficulty in measurement of this phase on better than ninety-five per cent of the subjects. The duration appears to be independent of heart rate (between rates 70–115), and inde-

pendent of age, sex and blood pressure. Ninety-seven per cent of the normals were found to have durations in the range of 0.060 to 0.140 seconds, inclusive. This measurement was taken on twenty-two patients with heart disease, consisting of myocardial infarcts, bundle-branch blocks, and hypertension. Significant increases in duration of this phase were noted in a majority of the cases. Eighty-six per cent were found to have durations in the range of 0.150 to 0.220 seconds, inclusive. This prolongation in isometric relaxation has not been found to be characteristic of any one pathologic condition studied. These results suggest that more importance may be given to this phase of the ventricular cycle, and further investigation into the physiologic factors concerned is indicated.

Effects of the ablation of the pituitary, adrenals and typhoid on liver regeneration, nucleic acid partition, etc. DAVID RAPPORT, ATTILIO CANZANELLI and RUTH GUILD (by invitation). *Department of Physiology, Tufts College Medical School, Boston.* Partition of ribosenucleic acid (PNA) and desoxyribosenucleic acid (DNA), total solids, total N, and the amount of liver regenerated were studied in rat livers before partial hepatectomy, in which about ⅔ of the liver was removed, and after 96 hours of regeneration. In all cases, the control and experimental animals received approximately the same amount of a synthetic diet. The findings below are to be construed as a comparison with results obtained on animals with intact endocrine glands. (1) *Pituitary* Hepatectomy was performed 4 to 7 days after hypophysectomy. In the pre-hepatectomy liver there was a small increase in PNA, but no other significant findings. In the regenerated liver, DNA was significantly increased, and the amount of liver regeneration was reduced to ½ of the control. (2) *Adrenals* PNA was increased both in the pre-hepatectomy liver (10 days after adrenalectomy) and in the regenerated liver. There was a tendency towards reduction in regeneration. When the animals were subjected to a temperature of 31°C. during the entire experiment, the decrease in regeneration became much more marked. (3) *Thyroid* Six weeks after thyroidectomy, the pre-hepatectomy liver showed a reduction in PNA and total N; the regenerated liver showed only tendencies which were not statistically significant. However, animals fed desiccated thyroid showed by comparison with the normal, and particularly with the thyroidectomized animals, a significant rise in PNA of both the pre-hepatectomy and the regenerated liver, as well as an increase in the amount of liver regenerated.

The results seem to indicate independently acting effects on the liver by the three glands studied.

The partial purification and some properties of an adaptation-stimulating principle from yeast. JOHN M. REINER and S. SPIEGELMAN. (*Department of Bacteriology and Immunology, Washington University School of Medicine, St. Louis, Mo.*) Recent studies of adaptive enzymes in yeast have suggested the hypothesis that enzymes are formed by self-duplicating cytoplasmic units. The existence in adapted cells of materials having some of the properties to be expected from such units was reported earlier (Cold Spring Harbor Symposia on Quantitative Biology, **11**: 256 (1946): J. General Physiology, **30**: 755 (1947). We now have a reproducible procedure by which an adaptation-stimulating principle can be prepared from galactose-adapted yeast cells, and separated from an as yet unidentified inhibitor. The active principle does not appear to be dialyzable. It is inactivated by brief exposure to cold acetone and by prolonged exposure to cold alcohol, but resists heating at 60°C. for as long as 45 minutes. Its activity is unaffected by desoxyribonuclease, ribonuclease, pepsin, trypsin, chymotrypsin, or papain. Incubation with protein denaturants such as urea inactivates the principle. The denaturants also inhibit normal adaptation; but the cells recover and eventually adapt. When the active principle is added to cells simultaneously with the denaturant, no inhibition is observed. Addition of the adaptive substrate prior to or together with the denaturant also has a protective effect. An important characteristic of this material is its extreme instability in the absence of the specific adaptive substrate. Usually all steps of a preparation are performed in the presence of the substrate (1% galactose). Omission of galactose or substitution of glucose leads to disappearance of activity. No desoxyribonucleic acid was detected in this material. Determinations of nitrogen, phosphorus, and ribose are consistent with the supposition that a nucleoprotein may be present. Prolonged treatment with chloroform to remove extraneous protein does not affect the activity, and the residue still contains large amounts of protein and has an N-P ratio too high for a nucleic acid. The possibility that the active material is the adaptive enzyme itself has been ruled out by experiments in which it was added to active extracts from unadapted yeast, which have been shown to possess all the non-protein components necessary for the adaptive fermentation (Archives of Biochem., **13**, 113 (1947)). The combination fails to ferment galactose.

Effects of hemorrhagic shock on hepatectomized and bilaterally nephrectomized, hepatectomized dogs. J. J. REINHARD, JR. (by invitation), O. GLASSER (by invitation) and IRVINE H. PAGE. *From the Research Division of the Cleveland Clinic Foundation Cleveland, Ohio.*

Hepatectomized and bilaterally nephrectomized-hepatectomized dogs were subjected to a standard hemorrhagic shock. The response of these animals did not differ significantly from that of animals

with livers and kidneys intact. When the dogs' own blood was therapeutically restored by intra-arterial transfusion the blood pressure increased and was maintained at the pre-shock level for 2 to 3 hours. Responses to adrenalin in the pre-shock control period and the post-shock control period were equal.

Other hepatectomized dogs were given 250–300 cc. of blood from animals in hemorrhagic shock whose livers were intact. In order to keep the blood volume constant, the blood from the shocked donor was transfused into one leg while an equal amount of blood of the hepatectomized dog was removed from the other leg. Some of the donor animals had intact kidneys and some had been bilaterally nephrectomized prior to shock. The hepatectomized recipients showed no toxic effects of the transfusion and the blood pressure remained constant.

To demonstrate further the lack of toxic effects following transfusion, the hepatectomized recipients were bled to produce shock. The response to bleeding was identical to that of hepatectomized dogs which had not received blood from a shocked donor.

Thus, it is seen that the absence of the liver does not change significantly the response of dogs to hemorrhagic shock.

Some cutaneous responses to "reflex heating." WALTER C. RANDALL, DOUGLAS E. SMITH, and ALRICK B. HERTZMAN. *Department of Physiology, St. Louis University, School of Medicine.*

Simultaneous observations were made upon the cutaneous blood flow, the skin surface temperature, and the number of functional sweat glands on the forehead, cheek, forearm, and finger pad while normal male subjects were exposed to "reflex heating" experiments. Immediately following exposure of the lower extremities and the lower trunk regions in a hot water bath (38–44), blood flow and skin temperature of the finger pad quickly increased to high levels. Simultaneous sweating responses on the finger pad showed either no change, or a slight increase. In all other areas studied a definite time lag followed the exposure to heat before increases in blood flow, temperature and sweating were observed. Increases in sweating and in blood flow generally occurred approximately together, although in some experiments the sweating responses appeared somewhat earlier than vasodilatation. Relative flow and sweating responses in the four body areas both in the normal and the heat dilated state, are described and compared with maximal responses induced by direct stimulation by histamine and mecholyl.

Physiological protection of the extremities from severe cold. S. I. RAPAPORT (by invitation), E. S. FETCHER, and JOHN F. HALL (by invitation). *The Aero Laboratory, Wright Field, Dayton, Ohio.*

Hand and foot temperatures in the cold can be made primarily dependent upon the state of thermal balance of the body; and nearly independent of ambient temperature and insulation.

Forced internal ventilation of clothing permitted control of the thermal state of the body through regulation of a hot air supply (see Fetcher, Rapaport, and Hall, this issue). The hands and feet were excluded from the ventilating circuit, receiving heat only from their blood supply. The insulation of the footgear and gloves, calculated in clo, was 2.6 and 0.9 respectively; that of the body clothing was 2.5 clo. The effect upon hand and foot temperatures of variation of the heat supplied to the body was studied at 0°F. and −30°F. It was found, in three subjects, that normal hand and foot temperatures were maintained when approximately 180 kg.Cal./hr. were supplied at 0°F., and 280 kg.Cal./hr. at −30°F.; *i.e.*, when the body did not need to conserve heat. The temperature of even the bare hand could be kept within the comfort range at −30°F. When the heat supply to the body was reduced, insulation could not prevent hand and foot temperatures from falling. In two of the three subjects, at −30°F., the extremities rewarmed after the body had been rewarmed. Hand temperature rose before foot temperature. Slight heat deficit caused a fall of foot temperature but not of hand temperature, despite the greater insulation of the foot.

Effect of epinephrine on the circulating eosinophils. L. RECANT (by invitation), P. H. FORSHAM (by invitation), and G. W. THORN. *Department of Medicine, Harvard Medical School, and the Medical Clinic, Peter Bent Brigham Hospital, Boston, Massachusetts.*

It has previously been shown that epinephrine activates the pituitary-adrenal cortical system by stimulating the secretion of adrenocorticotropin. It was subsequently demonstrated that adrenocorticotropin produced a fall in circulating eosinophils in subjects with normal adrenal cortical function. It has been found that 1.5 mgm. of epinephrine in saline given intravenously over a one-hour period produces a fall in circulating eosinophils of from 55 to 75 per cent in four hours in both normal subjects and patients with Addison's disease. The finding that the "adrenalin effect" on eosinophils is independent of the adrenal cortex has been confirmed by the study of normal, adrenalectomized and hypophysectomized rats. The mechanism of action has been studied.

Arterio venous blood sugar studies on the kidney of the eviscerated dog. ROGER M. REINECKE and PAUL J. HAUSER (by invitation). *University of Minnesota.*

The concentration of fermentable reducing substance, i.e., blood sugar, usually becomes higher in the renal vein than in the aorta of the dog from

which the liver, pancreas, spleen, stomach and intestine except the lower colon have been removed. This phenomenon is not prevented by stopping the secretion of urine through increasing the pressure in the ureter.

The effect of changes in cardiac "competence" on right auricular pressure. RICHARD A. REISS (by invitation) and JOSEPH R. DIPALMA. *Department of Physiology, Long Island College of Medicine, Brooklyn, N. Y.*

Opinion is divided as to the primary factor responsible for the elevated venous pressure encountered in congestive failure. One view relates it to decreased cardiac competence, while the other maintains that it results from the increased blood volume. Experiments in open chest cats in which cardiac competence is decreased by near lethal doses of quinidine intravenously, by electrically induced auricular fibrillation, or by topical application of mecholyl, cause rises in venous pressure (right auricle) of the low magnitude of 1 to 3 cms. of water. In competent hearts saline infusions as much as ¼ the total blood volume produce only temporary rises of venous pressure with return to normal values in two to five minutes. When increased blood volume is imposed on an incompetent heart in the same animal, the elevation of venous pressure is still transitory in nature. It is to be concluded that neither cardiac incompetence, increases in blood volume, nor a combination of the two can completely explain the sustained, markedly elevated venous pressures seen in congestive failure. In our experiments to date, prolonged rises in venous pressure were obtained only after the following conditions were produced sequentially: 1) a marked decrease in cardiac competence with quinidine, fibrillation and mecholyl; 2) an increase in blood volume by saline infusion; 3) a partial return of cardiac competence with epinephrine. It is suggested, therefore, that these three conditions, particularly a partial return of cardiac competence, are essential for the persistent elevation of venous pressure.

Prediction of the stroke volume of man from brachial pressure pulse values. JOHN W. REMINGTON, W. F. HAMILTON, CHARLES R. NOBACK (by invitation), and JAY J. GOLD (by invitation). *Department of Physiology, University of Georgia School of Medicine, Augusta, and Department of Anatomy, Long Island Medical School, Brooklyn.*

The volume-pressure relations, corrected for body size, of 48 human aortas, covering an age span from 8 to 89 years, were compiled. The series was divided into age groups, and into a group of hypertensives. *In situ* aortic lengths are the same for all groups. A definite trend for an increase in volume with ageing, and with hypertension, was noted, but the overlap between groups is large. There is no correlation between diastolic size and the volume uptake per unit pressure increase. Hence the variability of $\frac{\Delta V}{V \Delta P}$ is considerably greater than that of the simple ratio $\frac{\Delta V}{\Delta P}$. The latter is not significantly changed by ageing nor by hypertension.

Volume distensibility tables were constructed for the calculation of the stroke volume from the pulse contours of 68 intrabrachial pressure pulses, 60 of which were kindly supplied by Dr. André Cournand. Predicted stroke index differs from that given by the direct Fick procedure by an average of 17%. An estimation from pulse pressure alone gives an average discrepancy of 23%. The correction of pulse pressure for the changing arterial distensibility with pressure, lowers the average discrepancy to 18%. The size of the error, and the small improvement made by the use of the contour calculation over the latter procedure, indicate: 1) variation in systolic drainage plays a small part in the inaccuracy of the latter method; 2) that contour variations between brachial pulses is small; and 3) that the major inaccuracy of both methods lies in the variation in arterial distensibility of different individuals.

Does injury to an axon promptly induce altered excitability in its cell of origin? BIRDSEY RENSHAW and HERBERT ROSENBAUM (by invitation). *Department of Physiology, University of Oregon Medical School, Portland.*

Two types of experiment on lumbo-sacral motoneurons of pentobarbitalized cats have given no support to the hypothesis that injury to the axon of a neuron promptly induces a change in the excitability of its soma. In one series determinations were made of the size of 2-neuron-arc reflex discharges initiated by dorsal root stimulation and recorded electrically in the tibial nerve of the lower leg before and after section of the nerve four to six centimeters peripheral to the assembly of recording electrodes. In a second series microelectrodes were used to record the tibial nuclear action potentials that were evoked by antidromic motor volleys initiated by delivery of supramaximal shocks to the tibial nerve in the lower leg before and after section of the nerve below the ankle. Dorsal roots had been sectioned in all experiments. Controls established that section of the tibial nerve did not significantly affect the recording of centrifugal impulses in the first series, nor the size of the antidromic volleys in motor axons in the second. Within four to six minutes after section of the nerve there occurred only very small and inconstant changes in the responses used to measure somatic excitability. If any change is produced promptly as a consequence of nerve section, it is so small under the conditions of the experiments as to be difficult to investigate by the sensitive meth-

ods utilized and would appear to be of doubtful physiological significance.

Are there arterio-venous shunts in the kidney? FRANCOIS C. REUBI (by invitation), HENRY A. SCHROEDER, and ARNOLD H. WILLIAMS (by invitation). *Department of Internal Medicine and the Oscar Johnson Institute, Washington University School of Medicine, and Barnes Hospital, St. Louis, Missouri.*

Recently Trueta, et.al., using various techniques, demonstrated the probability of arterio-venous by-passes in the kidneys of rabbits. Attempts were made to examine this problem in the dog and man. In dogs renal blood flow was measured either by a rotameter in the renal artery or by the renal excretion of para-aminohippurate. The extraction ratio of PAH by the kidney was determined as well as simultaneous renal arterio-venous oxygen differences. After the intravenous injection of epinephrine, it was found that the renal arterio-venous oxygen difference decreased almost regularly three to eight minutes later. After ten minutes, it often rose to a higher level than before injection.

In human beings essentially the same methods were employed, renal venous blood being obtained by catheterization. After subcutaneous injection of adrenalin (0.5 mg.), the renal A-V oxygen difference decreased markedly a few minutes later in some but not in all cases. The extraction of PAH fell slightly about twenty minutes after injection, but this change was not simultaneous with the decrease in the renal A-V O_2 difference. From these results it can be concluded that renal arterio-venous by-passes may at times appear under some conditions, but are not of great importance in the total renal circulation.

An analysis of the effects of large volumes of isotonic saline on the circulation of severely hemorrhaged dogs. MONICA REYNOLDS (introduced by MAGNUS I. GREGERSEN). *Department of Physiology, College of Physicians and Surgeons, Columbia University.*

Hemorrhage was produced in 27 dogs by bleeding out and immediately returning 25% of the blood collected (Walcott, 1944). One hour later 0.9% saline was infused slowly (15% of body weight). Changes in the oxygen consumption, arterial and venous oxygen, cardiac output, plasma volume, blood volume, hematocrit and plasma protein were measured. Only four of the animals died. The dogs were thirsty, drank copiously, and excreted large volumes of urine. Plasma proteins and hematocrits shortly after the saline were lower than a day later, the plasma proteins increasing 1 gm. %. Eighteen hours after the infusion the plasma volume averaged 8 cc. per kgm. above the control value but the total blood volume was 15 cc. per kgm. below the control. Immediately after hemorrhage the cardiac output was reduced approximately 70%. The saline brought it up to control values and 18 hours later it was 5% above the control. After the bleeding the arterial oxygen concentration fell from 18 to 16 vols. %. After the infusion it fell to 9 vols. % and the following day it was 10 vols. %. The venous oxygen, though lowered from 14 to 4 vols. % by the bleeding, rose after the infusion to 6 vols. % where it remained. The circulatory changes induced by the saline apparently account for the therapeutic effects observed.

Uterine contractions effective in dilating the human cervix, recorded by the multichannel strain gage tokodynamometer. S. R. M. REYNOLDS, L. M. HELLMAN (by invitation) and P. BRUNS (by invitation). *Carnegie Institution of Washington, Department of Embryology, and Department of Obstetrics, Johns Hopkins University, School of Medicine.*

Using a multichannel strain gage tokodynamometer for recording patterns of uterine contractions in gravid women (Science, October 31, 1947), the essential characteristics of contractions which dilate the cervix have been determined. The uterus in the first stage of labor has three functionally distinct regions. The fungus always contracts rhythmically. The lower uterine segment shows no evidence of contraction. The mid-zone of the uterus contracts synchronously with the fundus at the start but it becomes progressively less active as cervical dilatation proceeds. The reason why the mid-zone becomes less active is because the contractions here are of progressively less intensity and of shorter duration than in the fundus. Consequently, the work of the fundus, as measured in ergs per unit area is far greater there than in the mid-zone. Deviations from this pattern have been seen in uterine inertia, prolonged labors, and false labor. In multiparous women and in precipitate labors, the fundus may be the only strongly contracting part of the uterus.

Nitrogen balances of dogs continuously infused with 50% glucose and protein preparations? C. MARTIN RHODE (by invitation), WILLIAM M. PARKINS and HARRY M. VARS. *Harrison Dept. of Surgical Research, Schools of Medicine, Univ. of Pennsylvania, Philadelphia.*

Solutions of 50% glucose and glucose-protein supplying 45–50 calories/kg./day were constantly introduced into the superior vena cava through an indwelling, plastic, capillary tube and an infusion apparatus which permitted free movement of the dog in a metabolic cage. Equilibration on glucose for 10 days or until urinary nitrogen reaches 100–110 mg./kg./day was followed by 6 day periods on different protein preparations in hypertonic glucose. Calories, nitrogen and fluid volume remained constant throughout. Nitrogen, about 120 mg./kg./day, was administered as casein hydroly-

sate alone; as casein hhdrolysate with macromolecular gelatin, in equal amounts; and as gelatin fortified with methionine and tryptophane.

Dogs so fed for 4–6 weeks appeared normally active and in good physical condition. The level of urinary nitrogen during these continuous infusions of 50% glucose at 45–50 calories/kg./day was comparable to that reported by others who fed protein-free diets containing 70–80 calories/kg./day by mouth. The nitrogen administered in these experiments was better retained than that injected in single daily doses by others. Average daily nitrogen balances in 4 dogs consecutively receiving glucose, casein hydrolysate as amigen, amigen with Knox P-20 gelatin, and amigen were −106, −15, −26, −6 mg./kg./day respectively. Continuing the study in two of these dogs, comparative values of amigen, fortified gelatin, and amigen were −1, −8, +3. Fifty percent of the intravenous macromolecular gelatin appeared in the urine as precipitable protein.

An instrument for the measurement of total thermal radiation in the environment. CHARLES H. RICHARDS (by invitation) and JAMES D. HARDY. *From the New York Hospital and the Department of Physiology, Cornell University Medical College, New York, N. Y.*

An instrument has been designed and built for the purpose of measuring the total thermal radiation under various environmental conditions. For example, in an outdoor environment this measurement is the sum of the direct radiation of the sun, the reflected radiation, the radiation exchange with the sky and with the terrestrial objects. It consists of two hollow spheres of silver, 6 mm. in diameter. Each contains a copper-constantan thermocouple soldered to the inside surface and a heating coil of approximately 50 ohms. The balls are supported by stainless steel, #17 hypodermic needle tubing through which are brought out the leads for the thermocouple and heater. Both the balls and the supports are plated with chromium on nickel. One ball is left polished for low emissivity, while the other is blackened with a coating having a high emissivity in the infrared spectral region. In any environment the temperature difference between the black ball and the polished ball will depend on the total radiation exchange between the balls and the environment. Thus, the amount of heat necessary to bring the two balls to the same temperature is a measure of the total thermal radiation. Under these conditions convection effects cancel. Temperature equilibrium reaches 95% in 2 minutes, and relatively rapid changes in the radiation load of the environment can be followed. Typical values of total radiation measurements on a summer day in New York City showed that at 9 AM, before the sunlight was directly incident on the balls, the radiation was minus 60 Cal/hr/m². In direct sunlight the total radiation increased until at 2:30 PM a measurement of plus 100 Cal/hr/m² was obtained. The radiation slowly decreased until shadows fell over the apparatus at about 6 PM and the radiation was minus 15 Cal/hr/m².

Pulmonary circulation during exercise in normal individuals and in patients with chronic pulmonary disease. R. L. RILEY (by invitation), A. HIMMELSTEIN (by invitation), H. L. MOTLEY (by invitation), H. M. WEINER (by invitation) and A. COURNAND. *Cardio-Pulmonary Laboratory, Chest Service, Bellevue Hospital, New York City, and the Department of Medicine, College of Physicians and Surgeons, Columbia University.*

Studies of cardiac output and pulmonary arterial pressure were performed using the venous catheter technic in three normal individuals and in eight patients with various types of chronic pulmonary disease. Measurements were made at rest and during exercise on a stationary bicycle. Two of the three normal subjects showed a decrease in the mean pressure in the pulmonary artery during exercise; all showed a marked drop in pulmonary vascular resistance and a minimal increase in the work of the right ventricle during exercise. Three of the patients with chronic pulmonary disease showed a significant elevation of pulmonary arterial pressure at rest, and in all eight cases the mean pressure increased during exercise. There was either no change or an increase in the pulmonary vascular resistance during exercise, and the work of the right ventricle was invariably higher than in the normal subjects at a corresponding work level. The findings indicate that the expansibility of the pulmonary vascular bed during exercise is limited in patients with chronic pulmonary disease. Anoxia may contribute to the elevation of pulmonary arterial pressure during exercise in those patients whose arterial oxygen saturation falls.

X-ray density changes in the heart recorded by the electrokymograph. GORDON C. RING, CATHARINE R. MICHIE (by invitation) and M. J. OPPENHEIMER. *Department of Physiology, Temple University School of Medicine.* When an electrokymograph (see G. C. Henny, B. R. Boone and W. E. Chamberlain; Am. J. of Roent. & Rad. Ther., Vol. LVII, 409, 1947) is placed over the middle of the fluoroscopic silhouette of the heart, it will produce curves which look much like cardiometer tracings. One might at first think that these records could be used to measure stroke volume of the heart. This, however, is not possible, since a large heart will have less change in thickness for a given stroke than will a small heart. Assuming that the heart is a sphere and that its total volume (walls plus blood) at the beginning of ejection is 700 cc., the ejection of 200 cc. of blood will result in a change in diameter of 1.16 cm., whereas if the starting volume is 500 cc. the change for a similar ejec-

tion will be 1.54 cm. On this basis, it should be possible if one knows the stroke volume and the amplitude of density changes, to work back and find out how much the heart has dilated.

In testing the above hypothesis, we have produced variations in the stroke of the dog's heart by vagal stimulation and have recorded cardiac density changes, ballistocardiograms and arterial pressures. These results were then plotted as amplitude of density change per cc. of output against the stroke or work of the heart. The curves showing small density changes during large change in stroke, we believe, indicate a heart which dilates very little and is therefore in good condition. Epinephrine has a direct effect upon the force of cardiac contraction and when this is given, in spite of the increased arterial pressure, the heart dilates less than during control periods. Our technique makes it possible to demonstrate this in the intact animal.

Inconsistencies between experimental and theoretical electrocardiography. JANE SANDS ROBB. *Department of Pharmacology, Syracuse University College of Medicine.*

Theory states that the QRS portion of the electrocardiogram reflects the asynchronous excitation of the surface of the heart through the surrounding volume conductor to the indirect leads. In our experiments the ventricles were enclosed in an insulating glass cardiometer; a rubber diaphragm fitted snugly into the a-v groove. The QRS deflections were reduced in amplitude but still apparent. These waves certainly did not reach the indirect electrodes through a volume conductor in contact with the ventricular surface. In other experiments the pulmonary circulation was interrupted and the right ventricular cavity filled with M/5 KCl. Soon afterward, the right ventricle ceased to contract. The electrocardiogram showed S-T shifts and brief QRS deflections in all three standard leads. Internal injury potentials are thought not to affect the indirect leads. Moreover, the theory that the QRS is the algebraic summation of asynchronous, long, oppositely directed, monophasic potentials from the two ventricles would demand a simple monophasic negative wave in this case. If the KCl had depolarized only the endocardial surface, one would expect to see some contraction on the right and a prolonged QRS due to syncytial spread through unaffected muscle from the left or to the right ventricle. Finally, when M/5 KCl was placed on the anterior surface or the apex of an exposed heart neither of which had any *direct* contact with a volume conductor S-T shifts readily occurred. Present theory would demand that only areas in actual contact with volume conductors contribute to the electrocardiogram. Thus, present electrocardiographic theory does not adequately explain many experimental observations.

The role of the *vasa nervorum*, especially in regard to "referred pain." JOSEPH THOMAS ROBERTS. *University of Arkansas School of Medicine, Little Rock, Arkansas, and Gallinger Municipal Hospital, Washington, D. C.*

The normally abundant blood supply of peripheral nerves was decreased, experimentally and in patients with occlusive vascular diseases. The level and severity of neurological changes were correlated closely with the level and degree of ischemia of the nerves. Ischemic nerves showed histological degeneration. It is concluded that the *vasa nervorum* play an essential role in maintaining the structure and function of peripheral nerves in a way similar to the role of the blood supply of the central nervous system. The blood supply of the autonomic nervous system seems to play a similar role, according to demonstration of its abundance. These findings are helpful in explaining: (1) variation in peripheral neurological complaints; (2) the infrequency of cardiac pain with cardiac hypertrophy and its frequency with other forms of myocardial ischemia; (3) "referred" pain. Somatic nerves of skin, muscle, or body wall may become painful due to vasospastic neural ischemia arising from stimuli with origin in viscera and with a pathway through autonomic nerves attached to the same or adjacent spinal segments.

Potentiation of the gastric secretory response to histamine by parasympathomimetic drugs. C. R. ROBERTSON (by invitation) and M. I. GROSSMAN. *Dept. of Clinical Science, Univ. of Illinois College of Medicine, Chicago.*

Both histamine and parasympathomimetic drugs stimulate acid secretion by the gastric glands. In the present experiments it has been shown that the response to these two agents acting simultaneously is greater than the sum of their independent actions. Prostigmine methylsulfate (0.5 mg. subcutaneously) and urecholine (0.01 mg. subcutaneously every ten minutes) produce very little or no acid secretory response in dogs with vagotomized pouches of the entire stomach. However when these drugs are given while the response to histamine dihydrochloride (0.0125 mg. subcutaneously every ten minutes) is in progress the rate of acid secretion is markedly increased. In the case of progstimine the acid production rose from 42.5 mg. per hour to 108.3 mg. per hour (12 tests on 6 dogs), with urecholine the corresponding figures were 89.9 mg. per hour with histamine alone and 221.1 mg. per hour with histamine plus urecholine (6 tests on 6 dogs). This potentiation of histamine stimulation by parasympathomimetic drugs has important implications for the understanding of the effect of atropine and the effect of vagotomy on the effectiveness of various gastric secretory stimulants.

Pretreatment against decompression sickness. TRUE W. ROBINSON (introduced by J. W. HEIM).

Physiology Branch, Aero Medical Laboratory, Wright Field, Dayton, Ohio.

A series of tests were performed on a group of 24 individuals to determine the best of several pretreatments before prolonged exposure (six hours) to high altitudes. Experiments were carried on both in an altitude chamber at 30,000 and 38,000 feet and in a B-17 airplane at 30,000 feet. At 38,000 feet in the altitude chamber performance was slightly better when the pretreatment consisted in breathing room air while resting rather than in breathing room air while exercising on a bicycle. Noticeable improvement in performance at altitude occurred when subjects breathed oxygen for an hour before ascent. Exercise with oxygen was slightly less beneficial as a pretreatment than rest with oxygen and certainly less popular. At 30,000 feet the same results were obtained. At this altitude exercise was performed in the chamber after ascent. Whereas exercise as pretreatment was slightly detrimental to altitude tolerance, deep knee bends at altitude materially decreased the tolerance. A comparison was made between performance at 30,000 feet in the altitude chamber and in the B-17 airplane. Pretreatment in both cases consisted of breathing oxygen for one hour while resting before reaching 30,000 feet. Deep knee exercises were performed at altitude both in chamber and plane. No significant difference in altitude tolerance occurred. If anything the men did better in the plane than in the chamber, perhaps because they were less bored. Besides indicating a preferable type of pretreatment, these results validate the use of the altitude chamber as a means of determining resistance to decompression sickness under actual flying conditions.

Further evidence for a temperature-sensitive mechanism in a poikilotherm, the turtle. S. Rodbard and F. Sampson (by invitation). *From the Cardiovascular Department, Research Institute, Michael Reese Hospital, Chicago, Illinois.*

We have previously reported evidence for the presence of a temperature-sensitive center in cold-blooded animals (Fed. Proc., **6**: 191, 1947). We showed that the relationship which is seen between the body temperature and the arterial pressure of these animals is destroyed by section of the spinal cord or by destruction of the brain. In our present experiments we have warmed or cooled the brain of the turtles by means of a silver wire introduced through a small trephine hole in the skull. Temperature change was produced by heat conduction from a reservoir of water in which the wire was mounted. When the wire was warmer than the brain, the blood pressure, as determined from the aorta by optical recording, increased about 6 to 10 mm. Hg. When the wire was cooler than the brain, a fall in pressure of similar magnitude was seen. These results strengthen our belief that a temperature-sensitive center is present in the brain of cold-blooded animals, and that this center functions to adjust the internal environment of the animal to changes in body temperature.

Sodium and chloride reabsorption. J. C. Roemmelt (by invitation), O. W. Sortorius (by invitation), and R. F. Pitts. *Dept. of Physiology, Syracuse University College of Medicine, Syracuse, N. Y.*

In studies on normal dogs, preliminary to assessing the effects of adrenal cortical hormones on electrolyte reabsorption, excretion and reabsorption of chloride were determined at plasma levels ranging from 110 to 157 millimols per liter. These levels were attained by infusing 3.5 per cent NaCl at 10 cc. per minute. Although in the massed data the reabsorption of chloride per 100 cc. of filtrate was constant within limits of $\pm 10\%$ of the mean, in individual experiments there was a tendency for the quantity reabsorbed to diminish at higher plasma levels. The rates of proximal and distal tubular reabsorption of chloride were calculated from these data according to the formula of H. W. Smith. The values for distal reabsorption were constant within $\pm 15\%$. The serum levels for sodium ranged from 135 to 176 millimols per liter. The quantity of sodium reabsorbed per 100 cc. of filtrate diminished as the serum level was raised. The distal tubular reabsorption, calculated as for chloride, was constant within $\pm 15\%$.

Oximetric determination of cardiac output in man. Albert Roos (by invitation), J. O. Elam (by invitation), J. F. Neville, Jr., (by invitation) and H. L. White. *Department of Physiology and Laboratory of Applied Thoracic Physiology (Department of Surgery) Washington University School of Medicine, St. Louis.*

Arterial-mixed venous blood oxygen differences have been obtained by a recording modified Millikan oximeter in combination with hemoglobin determinations. Alveolar gas tensions in equilibrium with right heart blood have been produced by one deep inspiration of 6% CO_2 in helium, followed by incomplete expiration to the outside. The breath is then held for several seconds, after which rebreathing of 400 to 500 cc. into a bag is permitted. The oximeter curve indicating oxygen saturation falls rapidly; 7 to 10 seconds after onset of fall a plateau, lasting 4 to 10 sec., is reached, which is taken to indicate equilibrium between right heart blood and alveolar air. The plateau is terminated by a second drop, indicating recirculation. During the plateau right heart blood is presumably passing through the lungs unchanged; work to verify this assumption is in progress. Arteriovenous differences, together with oxygen consumption determinations, permit calculation of cardiac outputs, which, in the sitting position, ranged from 2.69 to 2.91 liters per M^2.

Relation of basal metabolic rate to vasodilatation and vasoconstriction of extremities of normal subjects as measured by skin temperatures. GRACE M. ROTH and CHARLES SHEARD. *Section on Physiology, Division of Physics and Biophysical Research, Mayo Clinic and Mayo Foundation, Rochester, Minnesota.*

Maddock and Coller in 1933 first demonstrated a simple linear relationship between the skin temperatures of the great toe and the basal heat production per unit of surface area. In 1940 Sheard and Williams found a similar linear relationship, which seemed to be of a dual character, between the average skin temperatures of the toes and the basal metabolic rate. From a study of 150 observations of normal subjects a linear relationship of dual character was found between the skin temperature of the toes and the basal metabolic rates. The ages of the subjects ranged from nineteen to seventy years and the heat production per unit of surface area ranged from 30 to 50 calories per square meter per hour. After a control period of at least an hour at an environmental temperature of 25.5°C. (78°F.) when the skin temperatures and basal metabolic rates have been determined, vasoconstriction and vasodilatation of the extremities were produced by placing the subjects first in a cold room at 21°C. (69.8°F.) for an hour and then in a hot room at 30–31°C. (86–88°F.) for another hour. It was found that vasoconstriction took place at a more rapid rate and to a greater degree in the subjects with low basal metabolic rates than those with higher rates whereas vasodilatation took place more rapidly in the subjects with higher basal metabolic rates. Basal metabolic rates, therefore, should be considered in studies concerning vasodilatation or vasoconstriction as measured by skin temperatures.

Metabolic balances in the cold environment: I. Nitrogen and water exchanges. JAMES L. A. ROTH and JOHN A. FRANTZ (introduced by J. W. HEIM). *Physiology Branch, Aero Medical Laboratory, Wright Field, Dayton, Ohio.*

Metabolic observations were made to study the influence of a USAF emergency ration and a restricted water intake upon the nitrogen, water and energy exchanges in human subjects residing continuously for nine days in a cold environment, −32°C. Control studies included fasting for six days in the cold environment and subsistence upon the emergency ration at room temperatures. The design of an emergency ration presents a problem because of the limitation in weight and space allowance in aircraft. Survival for fasting is several times longer than for thirsting. Thus, the advantages of a ration must be appraised in terms of its cost to water balance. The ration provided 1948 calories per day, 318 gm. carbohydrate, 41.6 gm. protein (46 per cent from whole egg), and 56.6 gm. fat. Because of its high biological value, egg protein was used to reduce solute load upon the kidneys.

Body protein sparing contributed by the ration amounted to 70 to 80 per cent of the loss incidental to fasting. No evidence of azotemia, dehydration or hemoconcentration was observed. The greater daily loss of body water during fasting (490 cc. compared to 245 cc. with the ration) is attributed to a greater release of water from tissue catabolism. The minimum water intake required during the first three days of a fast was slightly less than with the ration; whereas, that required during the subsequent three days of fasting was significantly greater than with the ration. Under the conditions studied, water balance was enhanced during continued ingestion of the ration.

Production of anaphylaxis in guinea pigs with weakly antigenic protein hydrolysates. L. W. ROTH, R. K. RICHARDS and I. M. SHEPPERD (by invitation). *Department of Pharmacology, Abbott Laboratories, North Chicago, Ill.*

The importance of the total number of sensitizing injections and the duration of incubation period has been established in the course of testing several hundred guinea pigs with partial hydrolysates of protein. When these substances were administered according to the usual techniques described in the literature for strong antigens, doubtful or negative, anaphylactic responses were obtained. If, however the total number of sensitizing injections was increased, and the period between the last injection and the challenging dose was at least one month, antigenicity could be demonstrated much more regularly. If either factor was diminished, frequency and degree of positive reactions decreased. Doubling or trebling the injection speed favored the production of typical anaphylactic symptoms. Partial despeciation by the hydrolysis was demonstrated by cross-shocking experiments. Beef hydrolysate-sensitized animals showed anaphylactic reactions when challenged with hog hydrolysate, and vice versa. However, desensitization was not always complete, because in some animals surviving the first shock, the immediate injection of the original sensitizing material produced a second series of anaphylactic symptoms. Reactions were not demonstrable when similar materials were injected into non-sensitized animals, ruling out anaphylactoid or peptone-like shock. Antigens were not completely precipitable by potassium alum since activity was detectable both in the precipitate and supernatant fluid.

Rate of penetration of electrolytes into nerve fibers. MORTIMER A. ROTHENBERG (introduced by DAVID NACHMANSOHN). *Department of Neurology, College of Physicians & Surgeons, Columbia University, New York.*

The exchange of ions between the nerve fiber and its environment is of paramount importance in the

mechanism of nervous action. Although a few data exist concerning the ion content of the interior of the nerve cell, our knowledge as to the permeability of nerve membranes is limited and based mainly on indirect measurements. The permeability of ions and their rate of penetration have now been measured directly. Giant axon of Squids were exposed to sea water in which the usual ions were replaced by radioactive ones, for varying periods of time. The axoplasm was extruded and the concentration of ions determined with a Geiger counter.—It was found that potassium which is 18 times more concentrated inside than outside, penetrates into the interior against the concentration gradient at a rather high rate. Within 60 minutes, the concentration of radio-active ions was about twice as high in the cell interior as in the outside fluid. Sodium penetrates for the first 20 minutes of exposure at a high rate but then the process virtually stops in spite of the high outside and low inside concentration. At the end of 1 hour, the concentration inside is about one-fourth of that outside. Calcium behaves similarly to sodium although the ratio of concentration inside compared to that outside is higher than in the case of sodium. The implication of these findings will be discussed.

Progesterone or testosterone alter the human vaginal smear. BORIS B. RUBENSTEIN. *Department of Metabolism-Endocrinology Research, Michael Reese Hospital.*

There has been controversy whether the ovulation change in the human vaginal smear is due to the presence of progesterone or to the withdrawal of estrogen or combinations of the above hormonal factors. The estrogen-induced cornification of the human vaginal smear after castration is altered despite continuous estrogen medication within 24 hours after the injection of 25 mg. of testosterone propionate or of 40–80 mg., of progesterone. Discontinuing estrogen produces equivalent effects only after 3–5 days. The rapid smear change after progesterone suggests that the previously described "ovulation change" is indeed due to the simultaneous elaboration of both estrogen and progesterone, the latter leading to the rapid desquamation of the cornified epithelium.

Vitamin E diminishes the vasomotor symptoms of menopause. BORIS B. RUBENSTEIN. *Dept. of Metabolism Endocrinology Research, Michael Reese Hospital, Chicago.*

There have been many conflicting reports regarding the value of vitamin E in the treatment of menopause. Some of the discrepancies arise from the fact that 90% of women pass through the menopause without symptoms requiring medical attention, and of the remaining 10%, more than half may obtain adequate relief from small doses of barbiturates, or even from placebos. The present study used as subjects 17 patients with severe vasomotor symptoms (hot flashes) following surgical or irradiation menopause. These patients obtained little relief from barbiturates and none from placebos. All obtained prompt and complete relief from Dienestrol (Ortho Products Co.) (0.3-0.5 mg/da). In 14 of the 17 patients marked reduction of hot flashes (less than 1/da) occurred with 75 mg/da of vitamin E (Ephynl acetate, Roche). Only 6 of the 17 patients obtained complete relief from hot flashes with this medication.

The vaginal smears of all 17 patients showed minimal degrees of cornification after 30 days of medication, equivalent to the cornification produced by 25 mg., of testosterone propionate 3X/WK, however, vitamin E produces none of the masculinizing effects of testosterone. Vitamin E may be useful in the treatment of those cases of severe menopausal symptoms in which estrogens are contraindicated.

A comparison of heat-coagulable serum solids with serum protein determined by a Kjeldahl method. C. H. WILLIAM RUHE and PAUL L. MCLAIN (introduced by C. C. GUTHRIE). *Department of Physiology and Pharmacology, School of Medicine, University of Pittsburgh.*

The protein content of serum determined by a macro Kjeldahl method and the heat-coagulable, chloroform and water insoluble solids determined by the gravimetric method of Guthrie both vary directly with the specific gravity. Since the two measurements are closely related, a comparison of the methods was made on ten samples of beef serum and on ten known dilutions of each sample. The results showed mean values of 50.63 mg. protein per ml. of serum and 55.19 mg. solids per ml. of serum. In every case, the value for the solids was higher than the protein value. The difference between the groups was statistically highly significant. Analysis by dilutions to determine whether or not the differences varied with dilution showed that the mean per cent deviation of the solids from the protein varied from 10.32% to 6.95% and averaged 8.61% for all dilutions; there was no apparent relationship between the degree of dilution and the per cent deviation. When the methods were used to determine degree of dilution and applied to known dilutions of serum, they yielded mean errors of 6.1% for the Kjeldahl and 3.4% for the solid method. On this basis and from the standpoint of duplication of results, the solid method was more reliable.

Sympathetic reaction in peptic ulcer. A. H. RYAN. *Dept. of Physiology and Pharmacology, Chicago Medical School, Chicago, Illinois.*

The data consists of measurements of a sympathetic response to a standard stimulus. The standard stimulus was a ride of one minute on a bicycle ergometer, and the response, the palmar skin resistance (PSR) measured near the end of the

ride (Ryan and Ranseen, Am. J. Physiol. **142:** 68, 1944). A lower PSR indicates a greater sympathetic response and a higher PSR a lesser response. Approximately 500 daily measurements were made on 5 subjects with peptic ulcer (4 were students) over periods of 1.5 to 3 years. Two had hemorrhages; all had episodes of pain or other symptoms. Forty-five non-ulcer subjects (chiefly students), on whom 1360 daily measurements were made, served as controls. The mean PSR for the ulcer subjects was 3817, SD1278; and for the controls 2493, SD492. The difference is statistically significant. The following table summarizes the average PSR for periods with marked symptoms and periods with few symptoms, for the data available. The difference for each subject is highly significant statistically. Evidence of increased sympathetic response generally was present within the two-week period preceding symptoms.

Subject	Marked symptoms	Few symptoms	Difference
Ga (1)	2553	2938	385
Jo (2)*	5573	6834	1261
Br	3203	3763	560
Je	2246	2751	505

(1) 3 hemorrhages (2) 2 hemorrhages * female.

Some effects of stimulation of the orbital surface of the frontal lobe in the dog and monkey. ERNEST SACHS, JR. and SAMUEL J. BRENDLER (introduced by JOHN F. FULTON). *Laby. of Physiology, Yale Univ. School of Medicine, New Haven, Conn.*

Spencer (*Philos. Trans.*, 1894, *B*185: 609–657) demonstrated that respiratory arrest and blood pressure rise could be obtained by faradic stimulation of the orbital surface of the frontal lobe in cats, dogs, rabbits and monkeys. Bailey and Sweet (*J. Neurophysiol.*, 1940, **3:** 276–281) have observed similar respiratory arrest and blood pressure rise in the cat and monkey upon stimulation of the posterior part of the orbital surface. Delgado, Fulton and Livingston (*Fed. Proc.*, 1947, **6:** 95–96) find that certain foci on the orbital surface when stimulated cause an occasional instantaneous fall in blood pressure as well as respiratory arrest (dog and monkey). We have attempted to localize the focus in dogs and monkeys under Dial anesthesia, and our observations are in agreement with the above observers. We have obtained respiratory arrest in expiration and we also found it possible to inhibit respiration in any phase of its development. On the postero-medial portion of the orbital gyrus of the monkey is a circumscribed area of maximal sensitivity for respiratory arrest. The threshold for arrest rises upon moving away from this point in any direction, even as little as one millimeter. From this same area the blood pressure fall occurred almost immediately without change in pulse. Sharp rises in blood pressure were obtained by stimulation of the anterior perforated space in the monkey, using the same intensity and frequency of stimulus as was used for the examination of the orbital gyrus.

Extra-thyroidal iodine. WILLIAM T. SALTER, MACALLISTER W. JOHNSON (by invitation), and JANET BEACH (by invitation). *From the Laboratories of Pharmacology and Toxicology, Yale University School of Medicine, New Haven, Connecticut.*

In hyperthyroid organisms the organically bound iodine in serum, lymph and peripheral tissues is more concentrated than normal. In myxedema or marked hypothyroidism the concentration is very low. The relative changes, however, vary considerably in different species. For example, the skeletal muscle of the normal rat contains only approximately 6 micrograms percent (fresh tissue) as compared with 25 for the cat. When extracts of peripheral tissues are fractionated, the organically bound iodine can be separated into arbitrary fractions, associated with various protein constituents. In the case of skeletal muscle nearly all of the organic iodine is precipitable with the proteins of the tissue, whether the precipitation be made with salt (ammonium sulfate) or with organic solvents. The bulk of the organic iodine is precipitated with the muscle globulin ("myosin"), but relatively higher concentrations of iodine are found in the smaller moiety associated with the muscle albumin ("myogen"). These observations suggest that the organic iodine is not bound to the protein by simple adsorption, but rather is associated with a specific colloidal carrier.

Transverse latency relaxations of muscle stimulated with massive transverse shocks. ALEXANDER SANDOW. *Washington Square College of Arts and Sciences, New York University.* Initially tensed frog sartorii, immersed vertically in a curare-Ringer bath, are stimulated, isometrically, by transverse massive shocks impressed on two large, plane Ag-AgCl electrodes symmetrically flanking the muscle. Transverse mechanical latency changes are recorded by the piezoelectric, cathode-ray oscillographic method. The pickup stylus is an insulated, vertical, 9 cm. long #17 piano wire with a cross-bar at its free end which is pressed transversely against the muscle, any point of which may be studied by vertical pickup displacements. Due to the great stylus length, latency changes are distorted by the 0.5 ms. transmission time along the stylus and an initial slight force at the pickup crystal opposite to that impressed on the stylus end contacting the muscle. The second type of distortion is absent for stylus lengths less than 2.5 cm. (Thus, Schoepfle and Gilson's observa-

tion, made with a 5 cm. stylus, that a water-filled finger cot muscle model transmits a mechanical wave with an initial opposite phase is attributed to a stylus artifact, for it is not obtained with a short stylus.)

The actual experimental records, properly corrected for the sylus distortions, demonstrate that the first response at all muscle points is the latency relaxation, the latency of which is 1.0 ± 0.1 ms. throughout the pelvic ⅔ portion of the muscle, and about 1.3 ms. at the tibial end. Thus the results prove that the latency relaxation is a true physiological precontractile elongation of muscle, and that all portions of at least the pelvic ⅔ part are practically simultaneously excited by a transverse shock.

Observations on the thoracic wall respiratory reflex. D. Saris (by invitation), E. A. Reed (by invitation) and J. C. Scott. *Department of Physiology, The Hahnemann Medical College, Philadelphia, Pa.* This investigation differed from the procedure of Whitehead and Draper in: using single intravenous injections of pentothal to induce respiratory arrest, oxygen by endotracheal tube, stimulation of the chest wall by application of weights to a freely moving piston, recording of reflex responses by a Marey tambour connected to the tracheal catheter. Thirty-two experiments were performed, mainly on dogs using nembutal anesthesia. With injections of 1 cc. of 2.5% pentothal the average recovery rate of excitability of the respiratory center in terms of the strength of stimulus is roughly linear, a maximum stimulus being effective at about ½ the period of inhibition, a stimulus of ½ this strength at 75% of the period and a stimulus of ¼ this value at 95% of the total time. Subminimal stimuli produce relaxation of the abdominal wall. The duration of inhibition apparently depends upon the rate of destruction or elimination of pentothal and the rate of accumulation of CO_2. Bilateral cervical vagotomy markedly prolongs the period of inhibition and also abolishes the reflex. Section of the right vagus accounts for most of these effects. In one experiment the reflex was elicitable after section of the scalinus medius and rectus abdominis muscles and also by pressure on the parietal pleura in the region of the fourth interspace after the overlying structures had been removed. Present data suggests the reflex may be the Hering-Brewer type.

Changes in renal function in experimental metabolic acidosis in the normal human subject. O. W. Sartorius (by invitation), J. C. Roemmelt (by invitation) and R. F. Pitts. *Dept. of Physiology, Syracuse University College of Medicine, Syracuse, N. Y.* It has been repeatedly observed that early in the development of metabolic acidosis increased anion excretion is largely balanced by increased excretion of fixed base. Later, as the alkali reserve is depleted, increased ammonia excretion permits the conservation of fixed base. The changes in glomerular and tubular function which account for these alterations in fixed base excretion have been studied in a normal subject maintained on constant food and mineral intake for 15 days. From the 6th through the 10th day metabolic acidosis was induced by ingestion of ammonium chloride. Glomerular filtration rate and renal plasma flow were measured in 5 short tests performed at intervals during the control, acidosis, and recovery periods. Daily blood samples analyzed for chloride, sodium, pH, bicarbonate, and phosphate and 24 hour urine specimens analyzed for these same constituents, and in addition titratable acid and ammonia, permitted the description of the renal response in acidosis. The major findings follow. Filtration rate and renal plasma flow decreased during acidosis associated with the reduction in extracellular fluid volume. Excretion of chloride increased in consequence of an increase in the quantity delivered into the glomerular filtrate. In the initial stage of acidosis sodium reabsorption diminished despite reduced filtration, and as a consequence plasma concentration fell. Later with stimulation of ammonia production an increased proportion of the filtered sodium was reabsorbed and depleted reserves were conserved. During the recovery period a maintained high rate of ammonia excretion brought about a rapid restoration of the alkali reserve of the body.

Development of chronic hypertension in dogs by the occlusion of the carotid arteries. Ronald E. Scantlebury (introduced by T. L. Patterson). *Department of Physiology and Pharmacology, University of Arkansas School of Medicine.* In acute experiments blood pressure has been increased by ligation of the carotid arteries. To determine if similar results could be induced in chronic animals a series of experiments was instigated in which the common carotids of dogs were ligated. Blood pressure readings were taken from the hind legs of three dogs by means of a pediatrics cuff attached to a Baumonometer. The median of three hourly readings comprised the daily average. Normal blood pressure was determined daily for two months after which the common carotid arteries were occluded with wire sutures. Daily readings were continued during the week recovery period, followed by weekly readings. After operation on April 2nd Dog I, with a normal average systolic pressure of 129, showed a postoperative average of 166 with variations between 138 and 216. Dog II, with a normal average systolic pressure of 132, after operation on June 24th showed an average pressure of 169 with variations between 138 and 195. Dog III, with normal blood pressure of 115, after operation on June 26th showed average pressure of 152 with variations between 148 and

160. Due to the probability of error of the method employed an increase of at least 20 mm. Hg. was required before the animal was considered hypertensive. The average increase for each of three dogs was 37 mm. Hg. These animals were still living on December 6th. Studies to determine tissue changes which might indicate the mechanisms involved have not yet been made.

Blood electrolite changes in experimental renal hypertension. C. A. SCHAFFENBURG (by invitation) and H. SELYE. *Institut de Medecine et de Chirurgie expérimentales, Université de Montreal, Montréal, Canada.* The left kidney was transformed into an "endocrine kidney" in 24 female albino rats (averge weight, 150 gm.), using a previously described technic [Selye and Stone, J. Urol. **56:** 399 (1946)]. Post-operatively, they were maintained for ten days on "purina fox chow" and 1% NaCl solution ad lib. Previous observations, made in this Institute, revealed that under these conditions severe renal hypertension ensues. A similar group of unilaterally nephrectomized rats served as controls. After ten days both groups were fasted and given tap water during 24 hours; then they were bled by cutting the jugular vein and the blood was collected under oil in order to obtain serum for biochemical analysis. Seven animals of the first group whose kidneys proved (histologically) to have undergone perfect "endocrine transformation" showed marked hypochloremia (85.0 m.eq./L. ± 6.0) an increased CO_2 combining power (31 m.eq./L. ± 1.5) and a slightly decreased serum sodium (140.7 m.eq./L. ± 4.9). All these changes were statistically significant but there was no significant change in serum potassium (6.12 m.eq./L. ± 0.16). These findings are of interest especially since we found a similar (although more severe) hypochloremic alkalosis to be accompanied by hypokalemia in rats rendered hypertensive with desoxycorticosterone.

Electron microscope observations of nerve structure. FRANCIS O. SCHMITT and EDUARDO DE ROBERTIS (by invitation). *Biology Department, Massachusetts Institute of Technology.* Characteristic fibrous structures have been observed with the electron microscope in macerated preparations of all types of nerves thus far examined, vertebrate and invertebrate Most observations were made on formalin fixed material though the structures were observed also in macerated fresh frog sciatics and spinal roots. They were also obtained from fixed squid giant axons freed of non-axonic material. The chief features are the dense edges staining intensely with phosphotungstic acid, a core of relatively low density and an axial periodicity in the form of transverse bands. In human sympathetics the average width is about 600 A and the axial period is about 650 A; three intra-period bands of characteristic density and position have been observed. A dense amorphous material associated with the structures laterally is removable by water extraction.

The available evidence is consistent with this view that the fibrous axonic structures may be cylindrical or tubular in nature and are of indefinite length (at least many microns). They disintegrate very rapidly in macerated fresh material. In excised bull frog spinal roots kept in the cold the structures remain intact until the irritability is lost (8 to 10 days). Thereafter mainly disintegration products are observed. For these experiments action potential data were obtained, nerves fixed in formalin after various periods of survival and macerated preparations examined. Other details of structure of nerve fibers, as observed in the electron microscope, will be discussed.

Measurement of electrical energy release, "impedance", and longitudinal transport in nerve by differential electrode techniques. OTTO H. SCHMITT. *Departments of Zoology and Physics, University of Minnesota, Minneapolis, Minnesota.* By using closely spaced electrodes and high amplification instead of conventional nerve electrodes widely spaced or referred to a killed point, it is possible to measure directly the instantaneous axis cylinder current in single nerve fibers. Starting from this information and making free use of the new electronic techniques for differentiation and integration, it is easy to obtain directly on an oscillograph trace, measures of the electrical energy released at the nerve membrane during activity, of the generalized "Impedance"-source function which the membrane must represent, of the instantaneous charge deposited at the membrane, of membrane current, and even of the monophasic action potential. Introduction of the new concept of a generalized impedance-source complex to replace the separate conventional concepts of a polarizing "battery" and an A.C. impedance characteristic will be justified. Of special interest is the direct theoretical correlation found between the nerve after-potential distribution and the longitudinal movement of charge-bearing material in the axon.

Nerve excitation as a function of membrane voltage. GORDON M. SCHOEPFLE. *Dept. of Physiology, Washington University School of Medicine, St. Louis, Mo.* Excitation of frog nerve was investigated in relation to voltage changes on the surface of the nerve induced by threshold rectangular current stimuli of various durations. It is assumed that voltage across the membrane in the extrapolar region is proportional to the voltage of the external fluid in accordance with the cable theory of core conduction. For various durations of threshold current, curves of peak voltage as a function of extrapolar distance along the nerve surface were found to intersect at a locus corresponding to a region about

7 mm. distant from the cathode. In view of the very low magnitude of potential observed at such a great distance from the cathode it is concluded that these findings do not offer a direct measurement of the minimal length of nerve which must be raised to a critical level of excitation prior to initiation of a propagated response. In describing the time course of excitability at regions adjacent to the cathode a solution of the equation

$$\frac{dx}{dt} = C(y)(P_o - x) - k(x)$$

is found applicable as a first approximation. For this equation y is considered to be the concentration of a cation accumulated across the membrane capacity in direct proportion to the membrane voltage, and x is the fraction of a membrane constituent of original concentration P_o which combines with y to form a complex which when present in sufficient concentration produces instability of some membrane component and hence leads to response of the nerve.

Pressor substances in hypertensive blood. HENRY A. SCHROEDER, MELVIN L. GOLDMAN (by invitation) and NORMAN S. OLSEN (by invitation). *Departments of Internal Medicine and Biological Chemistry and the Oscar Johnson Institute, Washington University School of Medicine, and Barnes Hospital, St. Louis, Missouri.* Arterial blood from hypertensive and normal subjects was extracted according to the method of Schroeder and Stock (J. Clin. Invest. **21**: 627, 1942). Blood extracts were further purified by a method developed by one of us (N. S. O.). Blood pressure in the rat was measured directly by a Hamilton manometer, and the effect of the intravenous injection of blood extracts was determined. The crude extracts contained substances producing three effects, i.e. depressor, pressor, and one resulting in bradycardia. Purified extracts contained only depressor or pressor substances. The latter were not observed in extracts taken from the blood of normotensive subjects. Most hypertensive individuals contained in their blood pressor material which also gave a VEM reaction in the vessels of the rat's mesoappendix. Further attempts to isolate, purify and characterize this substance will be discussed.

Relation between growth and synthesis of lipid in colpidium. GERALD R. SEAMAN (introduced by CHARLES G. WILBER). *Biological Laboratory, Fordham University. Colpidium capylum* was grown in sterile, fat free proteose peptone solution and the amounts of lipids produced were estimated by standard biochemical procedures. The results were correlated with the growth phase of the respective cultures. Cholesterol was not found at any time in the cultures or in the colpidia. Table 1 shows the amounts of fatty acids present in the organisms in relation to the population of the cultures. Lipid phosphorus remains at a constant value of .0003–.0005 mg./1000 organisms. During the logarithmic phase of growth the amount of lipid phosphorus in the culture fluid remains at a constant level of 6.1–6.8 mg./100 cc. The fatty acids in the culture fluid at the beginning of the logarithmic phase decrease in amount from 21.5 mg./100 cc. to 13.9–15.2 mg./100 cc. and remain at that low level until the equilibrium phase of growth is reached.

Age of culture	Populations in 1000 organisms	Mg. fatty acids/1000 organisms
days		
2	13.2	.0022
3	23.1	.0007
4	57.3	.0020
5	61.6	.0014
6	62.9	.0010
7	59.8	.0016
8	53.8	.0012

The amounts of lipid phosphorus and of fatty acids in the culture fluid increase at the beginning of the equilibrium phase and continue to increase during the declining phase of growth of the culture.

Fetal gastric secretion of radioiodine applied precutaneously to pregnant animals. W. A. SELLE and OTIS B. MILLER (by invitation). *Departments of Physiology and Dermatology, University of Texas, Medical School, Galveston.* Iodine 131, in ointment or aqueous solution, was applied to the denuded, unbraised skin of pregnant rabbits and guinea pigs near term over an area of approximately 20 sq. cm. Following intervals of 12 to 24 hours the animals were sacrificed and the fetuses removed from the uterus with care to prevent contamination by the mother's skin, blood, urine. Samples of the amniotic fluid, urine, blood and of most of the organs were taken for examination by Geiger Counter technics. The distribution of radioiodine in the fetus was found to be quite similar to that of adult animals. While the fetal thyroid took up large amounts of the precutaneously transmitted iodine, more than 90% of the total fetal iodine was located in the gastric contents. The lung, kidney, and urine contained relatively high percentages, whereas the blood, liver, spleen, bile and submaxillary gland contained intermediate amounts. Amniotic fluid, muscles, skin, and intestine contained low percentages. The mechanisms of secretion and concentration of iodine by the stomach were not determined. It is thought that the iodine is secreted in a manner similar to that of chloride.

Further studies concerning brain lesions induced by desoxycorticosterone overdosage in the rat. HANS SELYE, PIERRE HAOUR (by invitation) and CLAUDE FARIBAULT, (by invitation). *Institut de Médecine et de Chirurgie Expérimentales, Université de Montréal, Montréal, Canada.* Previous investigations have shown that in addition to nephrosclerosis, hypertension and periarteritis nodosa,

overdosage with desoxycorticosterone acetate (DCA) can produce severe morphologic changes in the brain (Selye et al. Exper. Med. and Surg. **2**, 224, (1944); Selye, J. Clin. Endo. **6**: 117 (1946)). In continuation of this work 20 young rats (72 gm.) were given subcutaneous implants of three 20 mg. DCA pellets following sensitization to the toxic effects of the latter by unilateral nephrectomy. All animals were kept on "purina fox chow," but received 1% NaCl, instead of tap water, as a drinking fluid. During the course of the experiment it was noted that several of the animals showed marked neurological manifestations such as convulsions, motor disturbances in the fore-limbs or signs of general excitement and confusion. They also revealed a typical skull deformity, characterized especially by the enlargement and rounding of the calvarium, similar to that seen in the spontaneous hydrocephalus of the rat. After 43 days of treatment, at autopsy, these animals exhibited an increased average brain weight, multiple petechial hemorrhages under the surface of the hemispheres, an increased amount of cerebrospinal fluid and certain histologic changes in the nervous tissue which will be described in detail.

Measurement of red-cell volume loss by the use of methemoglobin-tagged cells. O. W. SHADLE and JAMES C. MOORE (introduced by HAMPDEN C. LAWSON). *The Dept. of Physiology, Univ. of Louisville School of Medicine.* The total circulating red-cell volume was estimated in barbitalized, splenectomized dogs before and after hemorrhage by the methemoglobin tagged-cell method of Moore and Shadle (Fed. Proc. **7**: 82, 1948). When arterial pressure was maintained by injecting gelatin solutions after the hemorrhage, good agreement was obtained between the first circulating cell volume and the sum of the post-hemorrhage volume and the volume of cells drawn. In a series of six dogs, the agreement between these two values was always within 4 per cent, with random directions of disagreement. In five additional experiments, mean arterial pressure was stabilized between 30 and 50 mm. Hg, after the hemorrhage, by additional bleedings or re-injections, before making the second determination. The post-hemorrhage volume, plus the cell-volume drawn, was significantly less than the first circulating volume in only one dog, which suggests trapping of cells at these levels of arterial pressure. In the remaining animals, although the reduction in cell volume measured by the second determination was always slightly excessive, deviation from the expected values did not exceed the variations observed when arterial pressure was maintained.

The renal clearance of some polyethylene glycols in the dog. C. BOYD SHAFFER, FRANCES H. CRITCHFIELD and CHARLES P. CARPENTER (introduced by J. M. ROGOFF). *Chemical Hygiene Fellowship Mellon Institute, Pittsburgh, Pennsylvania.* The polyethylene glycols (PEG) are a series of unctuous, water-soluble compounds varying in consistency from viscous liquids to pasty or solid materials. Structurally, they are characterized by a large number of ether linkages together with terminal hydroxyl groups, and they possess a low order of chemical reactivity. In this study the renal clearance rate of several PEG ranging in average molecular weight from 400 to 6000 have been investigated in the dog using creatinine as the reference substance. Determinations of PEG were made by a gravimetric method (Shaffer and Critchfield, Anal. Chem., **19**: 32, 1947).

Experiments were performed on female dogs lightly-anesthetized with pentobarbital. The PEG was administered in saline solution in concentrations up to 5 per cent by constant intravenous infusion at a rate of about 2.4 ml./min. A total of 59 periods were obtained in 16 experiments on PEG of average molecular weights of 400, 1000, 1540, and 4000. Taking these 59 periods as a group, the average ratio of the PEG clearance to the simultaneous creatinine clearance was 0.99 with a standard deviation of 0.06.

The ratio of the clearance of a PEG of average molecular weight 6000 to that of creatinine was consistently and significantly less than 1. This circumstance was essentially unaffected by extreme hyperglycemia and by the intravenous infusion of diodrast in an amount sufficient to saturate tubular excretory activity.

Injected intravenously, the PEG are distributed in a fluid volume approximating that of the extracellular fluid of the body.

Medium concentration in relation to the water content and electrical properties of nerve. ABRAHAM M. SHANES. *Department of Physiology and Biophysics, Georgetown University School of Medicine, Washington, D. C., Marine Biological Laboratory, Woods Hole, Mass., and Bermuda Biological Station. Rana.* Frog sciatics decrease in weight to 85 ± 1.5% in Ringer made 2X hypertonic by the addition of either KCl or NaCl; sucrose is significantly more effective, equivalent amounts (according to freezing point data) reducing the weight to to 78.6 ± 0.62%. Hypertonicity with sucrose decreases the resting potential but increases the action potential, while excess NaCl, applied to the entire trunk, reduces the action potential reversibly. Weight changes were completely reversible after ca. 20 hours in hypertonic solutions. *Libinia.* Spider crab leg nerves are freely permeable to KCl. Thus, the same degree of weight increase results whether distilled water or "isotonic" KCl is used to dilute the artificial sea water medium. Sucrose, glucose, and NaCl are less effective, in the order given, in causing weight loss when added in equivalent amounts to sea water to make a 1.5X hypertonic medium. The addition of KCl to the medium

has no prolonged, effect on the weight, although in other invertebrate nerves a reversible increase occurs. Hypertonic solutions decrease the resting potential, but conduction of the motor fibers is not prevented even in 2X solutions of NaCl. *Loligo*. Tonicity changes of the order 25–50% render the squid giant axon irreversibly inexcitable. Microscopic observation suggests that the sheath limits swelling in hypotonic sea water; in very hypertonic solutions (5X) the axoplasm pulls away from the sheath. No osmotic change occurred in isotonic KCl for as long as ca. 15 minutes, and excitability was restored upon return to sea water.

The alteration in osmotically inactive fraction produced by cell activation. HERBERT SHAPIRO. *Laboratory of Physical Biology, National Institute of Health, Bethesda, Maryland.* Resting and activated eggs of the sea urchin, Arbacia punctulata, were swollen in hypotonic sea water (60, 70, 80, and 90 per cent), and allowed to attain equilibrium, after which volumes were determined from filar ocular micrometer measurements of their diameters. Both fertilized and unfertilized eggs obey the Boyle-van't Hoff law ($P(V - b) = K$), but the value for "b", the osmotically inactive fraction (o.i.f.), or non-swellable volume was different for the two, averaging in the cases studied, 7.3 per cent for unfertilized, and 27.4 per cent for fertilized. On activation in normal sea water, the eggs of the sea urchin undergo a definite increase in total cell volume of approximately 4 per cent. Evidence is available that the alteration in o.i.f. and in cell volume may depend upon the species in question. In two species a parallelism between change in "b", and alteration of respiratory metabolism on fertilization was found, though this may not prove to be a general rule. Equations for the calculation of the point at which osmotic pressures and cell volumes are identical for unfertilized and fertilized are included. A mechanical analogue of the phenomena is introduced.

The retarding action of vitamin C in physiological concentrations on the rate of cell division. HERBERT SHAPIRO. *Laboratory of Physical Biology, National Institute of Health, Bethesda, Maryland.* A study of the pathological changes in ascorbic acid deficiency indicates that the primary effect is an abnormality in the intercellular material or perhaps an effect at the cell surface, resulting in changes in the bones and small blood vessels. The phenomena are of cellular physiological interest, as they point to some relation between cell structure and pathology, arising from a biochemical deficiency. Ascorbic acid apparently plays no major role in tissue respiration, hence it appeared promising to investigate the influence of l-ascorbic acid on cell division, as possible evidence of action on the cell surface. Fertilized eggs of the sea urchin, Arbacia punctulata, were exposed at constant temperature to concentrations of vitamin C in sea water, varying from 2.62×10^{-6} molar to 1.57×10^{-3} molar. The rate of cell division was determined by measuring the time required for 50 per cent of the cells to pass through first cleavage. Beginning with approximately 10^{-4} molar solutions, a definite retardation of cell division was observed. At high concentrations, complete inhibition of division occurred. The slowing down of the rate of cell division appeared to be roughly a linear function of the concentration, beyond a minimal effective concentration.

The effect of "carbonation" on the rate of gastric emptying and intestinal absorption of sucrose solution. K. L. SHAPIRO (by invitation) and A. C. IVY. *Dept. of Clinical Science, Univ. of Illinois College of Medicine, Chicago.* Ten healthy adult male subjects beween 24 and 32 years of age, were each fed after a 12 hour fast, 200 cc. of an uncarbonated solution of 10% sucrose in distilled water. The fluid volume and sugar content of this amount of solution are approximately equal to those of the average carbonated "soft-drink" beverage widely consumed to-day. Venous blood samples for quantitative sugar determinations were drawn just prior to the administration of the solutions and at intervals of ½, 1, 1½, 2, and 3 hours after its ingestion.

On another occasion, under the same conditions, and after a similar 12 hour period of fasting, the same ten subjects were each fed 200 cc. of a carbonated solution of 10% sucrose in distilled water. Venous blood samples for blood sugar estimation were drawn at intervals equal to those of the previous period of observation. The carbonation had no significant effect.

Effect of phytate and other food ingredients on the absorption of radioactive iron. LEON M. SHARPE (by invitation), ROBERT S. HARRIS, WENDELL C. PEACOCK (by invitation), and RICHARD C. COOKE (by invitation). *Departments of Food Technology and Physics, The Massachusetts Institute of Technology and the Walter E. Fernald State School.* Fifteen boys, 12 to 17 years old, living in an institution, were fed breakfasts of the following test meals: I, 200 ml. water; II, 200 ml. milk; III, 200 ml. milk plus 234 mg. phytic acid as the sodium salt; IV, 200 ml. milk, plus 234 mg. phytic acid as in rolled oats (285 gms.); V, 200 ml. milk, plus 285 gms. rolled oats, 150 ml. tomato juice, 75 gms. egg omelet and 56 gm. white bread. Total iron content of all meals was equalized with V at 8.31 mg. using $FeCl_3$. Ascorbic acid (32 mg.) added to reduce all iron to ferrous form. I and II contained no phytate. Fe^{55} and Fe^{59} were used alternately. No food was eaten before or until five hours after the test meals. Radioactive iron absorption was calculated from activity of hemoglobin, plasma volume and hematocrits. Maximum radioactivity of the hemoglobin was reached within 10 days after feeding. Average percentage of ingested iron appearing in the hemo-

globin was: I, 12.4; II, 7.7; III, 0.8; IV, 4.8 and V, 2.5. Sodium phytate markedly affected iron absorption and is five times more reactive than that in oatmeal. Milk and milk plus oatmeal decreased iron absorption by ⅓rd and ⅔rd, respectively. Thus phytate-free milk interfered with iron absorption nearly as much as phytate-rich oatmeal.

Adenosinetriphosphatase and conduction of the nerve impulse. WYNNE SHARPLES (by invitation), HARRY GRUNDFEST and DAVID NACHMANSOHN. *Dept. of Neurology, College of Physicians & Surgeons, Columbia University, New York.* Adenosinetriphosphatase has been found in high concentrations in the giant nerve fiber of *Loligo pealii* after the major part of the axoplasm has been extruded (Libet, Biol. Bull., **93**: 219, 1947). On the basis of this finding, that adenosinetriphosphatase like cholinesterase is concentrated in the surface of the fiber, Libet suggested that permeability changes of the nerve membrane associated with electrical activity may in turn be closely associated with adenosinetriphosphatase activity. This hypothesis has been tested by *in vivo* and *in vitro* experiments on frog sciatic nerves, by the use of anticholinesterases that abolish electrical activity rapidly. Eserine abolishes conducted electrical activity of nerve reversibly. It does not affect adenosinetriphosphatase activity even in 10^{-3} M concentration whereas cholinesterase activity is abolished in concentrations of 10^{-5} to 10^{-6} M. Similarly, di-isopropyl fluorophosphate which abolishes the electrical activity reversibly or irreversibly depending upon controlled experimental conditions, does not affect activity of adenosinetriphosphatase at a concentration of 10^{-3} M whereas it abolishes cholinesterase activity in concentrations of 10^{-5} to 10^{-6} M.

On the use of venous pressure in the head as a tolerance index of negative "G" in humans. R. S. SHAW (by invitation), J. L. GAMBLE, JR. (by invitation), P. J. MAHER (by invitation), J. P. HENRY and O. GAUER (by invitation). *Aero Medical Laby., Air Materiel Command, Wright Field, Dayton, Ohio.* Injury to animals from negative acceleration of over three seconds' duration first appears as rupture of small vessels about the head; consequently, it was thought that venous pressure in the head might be used as an index of tolerance in humans exposed to negative acceleration on the centrifuge. Four subjects were secured in a seat with an adjustable back mounted on the centrifuge, and venous pressures during negative acceleration were measured with a variable inductance manometer. Venous pressure in the frontal vein of human subjects exposed to negative acceleration was found to vary linearly with the magnitude of the acceleration as though from a simple hydrostatic column based at the heart; they ranged from 80 to 110 mm. Hg. No appreciable movement of the heart was noted in X-rays of the chest taken under negative accelerations of 3 g.; "Red-out" was not observed in this series. Pressures were recorded with subjects in 45-degree and 60-degree backward tilted seats, and compared with those obtained in the standard 10-degree seat tilt. No significant protection against negative acceleration as indicated by venous pressures in the frontal vein resulted from this tilting. Breath holding and straining maneuvers, which increase venous pressure in the resting subject, enhanced the disagreeable subjective sensations, while closing the glottis and attempting inspiration (Müller Maneuver), which decreases venous pressure, resulted in considerable alleviation of symptoms under negative accelerations up to 3 g.

Courses of dark adaptation and levels of vitamin A and carotene in normal and clinical conditions. CHARLES SHEARD. *Division of Physics and Biophysical Research, Mayo Foundation and Mayo Clinic, Rochester, Minnesota.* This presentation is concerned with tests on the levels and courses of dark adaptation of rods and cones, on the one hand, and, on the other hand, the levels of vitamin A in the blood serum in a group of seventy persons clinically selected because of a history or symptoms indicative of possible avitaminosis A. Standards, criteria and controls used in the determination of dark adaptation levels are those established by the author. Standards regarding high, average and low values of vitamin A (I.U.) and carotene (I.U.) per 100 cc. of blood serum are those established by Osterberg and his associates. In 60 per cent of the group tested, data show high threshold levels and delayed recovery of both rod and cone dark adaptation with very low fasting levels of carotene and vitamin A and with levels of vitamin A less than 500 I.U. per 100 cc. of blood serum six hours after ingestion of 7,500 units of vitamin A per kilogram of body weight. From the data the following conclusions are drawn: 1. The courses and the levels of dark adaptation are not in agreement with the normal (standard) findings when the fasting levels of carotene and vitamin A are very low and are less than 100 I.U. and 150 to 200 I.U. per 100 cc. of blood serum respectively, or, again, less than 500 I.U. per 100 cc. of serum six hours after the ingestion of 7,500 units of vitamin A per kilogram of body weight. 2. In some cases the levels of dark adaptation are elevated 0.5 to 1.0 log unit for the cones and as much as 1.5 log unit for the rods. 3. Definite cone knees (plateaus) are exhibited in the courses of dark adaptation in peripheral areas; these effects persist for as much as ten minutes and are produced under conditions of previous light adaptation which would not cause such results in persons with normal dark adaptation and high vitamin A and carotene levels in blood serums. 4. In perimetric surveys made by the technic of dark adaptation used by the author, it has been possible to show marked reductions in the functioning of the rods in certain areas

and, in some instances, marked improvement therein subsequent to vitamin A therapy or other medical treatment.

Inhibition of sweating by means of iontophoresis. WALTER B. SHELLEY, PETER N. HORVATH (by invitation) and STEVEN M. HORVATH. *Departments of Dermatology and Physical Medicine, University of Pennsylvania School of Medicine.* After treatment of areas of normal human skin with iontophoresis, in which distilled water, methylene blue solutions, and other compounds were used under the positive electrode, sweating was found to be inhibited. The skin appeared normal when the subject was not sweating but when profuse sweating was induced, multiple vesicles appeared at the sites of sweat gland openings. The inhibition of sweating was present immediately after treatment in some subjects, but the vesicles could not be made to appear until after a variable latent period. The effect was not permanent since normal function of the sweat glands returned after one to three weeks.

The permeability of normal human skin to histamine. WALTER B. SHELLEY and FRANK M. MELTON (by invitation). *Department of Dermatology, University of Pennsylvania School of Medicine.* A study has been made of certain of the factors influencing the penetration of histamine through normal intact human skin. The skin was found to be permeable to histamine both in the form of the salt and the free base. However, histamine free base penetrated the skin more readily than the histamine diphosphate. A second important factor was found to be the concentration of histamine which dissolved in the vehicle. Uniformly, as the concentration of histamine in solution increased, the magnitude and duration of effect increased. Solutions of 0.1% strength had no effect on the skin whereas 1% solutions (calculated as free base) produced local erythema, urticaria and pruritus. Water proved to be among the more effective of 21 semisolid and liquid vehicles studied. In the case of liquid vehicles, covering the inunction site greatly prolonged and increased absorption by preventing evaporation. Individual differences in the permeability of the skin to histamine were striking but in all subjects the application of a 10% aqueous solution of histamine free base produced marked local erythema, urticaria and pruritus.

The failure of certain previous investigators to demonstrate the permeability of the skin to histamine appears to stem from the fact that the histamine salt was used in low concentrations and in vehicles which did not promote absorption.

The composition of polysaccharides of human cervical mucus. LANDRUM B. SHETTLES and ZACHARIAS DISCHE (by invitation). *Departments of Obstetrics and Gynecology, Anatomy and Biochemistry, College of Physicians and Surgeons, Columbia University.* The presence of substances reducing after hydrolysis in human cervical mucus was reported by Viergiver and Pommerenke and of glucosamine in bovine cervical mucus by Boyland. We investigated the nature of polysaccharides in the translucent, acellular, midcycle human cervical mucus with the cystein reaction of carbohydrates described by Dische, which enables one to detect individual hexoses and various classes of true sugars in mixtures and which was adapted by us for quantitative purposes. The total amount of true sugars in the mucus ascertained by the indole method of Dische and Popper varied between 0.120 and 0.250%. Up to 20% of it consisted of methylpentose and up to 55% of galactose. The rest was glucose. No significant amounts of hexoketoses, mannose or hexuronic acids were found. The ratio: galactose/methylpentose varies between 2.5 and 2.9. This ratio appears identical with that found with the same and other methods in various preparations of blood group substances isolated by Kabat and collaborators from linings of stomach mucosa of hogs. The methylpentose and galactose of the mucus are precipitated by 90% ethanol. These findings suggest that the main bulk of polysaccharides of human midcycle cervical mucus consists of a substance identical with or closely related to one of the blood group substances.

Observations on the sustained pressor principle in different animal species. ROBERT E. SHIPLEY and O. M. HELMER. *Lilly Laboratory for Clinical Research, Indianapolis General Hospital.* Blood plasma obtained from dogs and rats which had been subjected to ½–2 hours of hypotension (40–50 mm. Hg) possessed the ability to cause a sustained elevation of blood pressure when injected intravenously into dogs and rats which had been bilaterally nephrectomized 1–3 days before. Plasmas from normal dogs or rats did not cause sustained pressor responses. These findings parallel the observations originally made on cats (Shipley et al., Am. J. Physiol. **149:** 708, 1947).

Semi-crude extracts of cat, dog, sheep, and hog kidneys were injected intravenously into cats and dogs and the animals bled 1 hour thereafter. The plasmas so obtained possessed the ability to cause sustained pressor responses in nephrectomized cats, dogs, and rats.

Experimental displacement of the acid base equilibrium of the blood in aged males. NATHAN W. SHOCK and MARVIN J. YIENGST (by invitation). *Division of Physiology, National Institute of Health, Bethesda, Maryland, and Baltimore City Hospitals, Baltimore, Maryland.* Experimental displacement in the acid base equilibrium of the blood were induced in six males, aged 70 to 90 years by oral administration of sodium bicarbonate or ammonium chloride. Blood samples were drawn at ½ to 1 hour intervals for 9 hours after administration of the salt, and analyses for pH_s, V_c, total CO_2 con-

tent, pCO_2, and bicarbonate content of the plasma were made by the Shock-Hastings micro method; blood samples were also drawn at 24, 28, 32, 48, 52, and 56 hours when necessary to obtain normal values. Results on the aged were compared with those previously reported on young adults (Shock, N. W. and Hastings, A. B., J. Biol. Chem., **112**: 239–252, 1935). Acid base paths of displacement and recovery and rates of recovery of plasma bicarbonate content were plotted for each experiment. Preliminary results indicate that recovery from metabolic acidosis induced by the oral administration of ammonium chloride is significantly slower in the aged subjects tested than in the young adults. Eight hours after the administration of 10 gms. of ammonium chloride, the average recovery of blood bicarbonate levels was 70 per cent complete in the young subjects, while in the old subjects the average recovery was only 27 per cent complete. The rate of elimination of orally ingested sodium bicarbonate was only slightly slower in aged males than in young adults. It was also observed that the respiratory compensation for a metabolic alkalosis observed in young adults did not occur in the aged subjects when 20 grams of sodium bicarbonate were administered orally.

Hepato-renal factors in circulatory homeostasis. XV. Vasotropic content of blood in chronic hypertension (dogs, man). EPHRAIM SHORR, B. W. ZWEIFACH and S. BAEZ (by invitation). *Dept. of Medicine, Cornell University Medical College and The New York Hospital, New York City.* During the development of renal hypertension (Goldblatt clamp) in the dog, blood samples, assayed by the rat mesoappendix test, produced a vaso-excitor effect on the terminal arterioles and precapillaries. Later, with the development of chronic hypertension little or no VEM could be detected in the blood stream by this method. This "neutral" reaction was found to represent a counterbalancing of the augmented VEM titer by a comparable increase in the oppositely acting VDM principle. This was demonstrated by aerobic incubation of "neutral" blood samples with normal kidney slices *in vitro*, a procedure which results in the inactivation of the VEM and thereby reveals the absolute concentration of VDM in the blood. Blood samples taken from patients with chronic essential hypertension and adequate renal excretory function were likewise found to produce a "neutral" effect in the test rat. The question arose as to whether the "neutral" effect in the blood of man represented a similar state of equilibrium with high titers of both VDM and VEM. This was conclusively demonstrated in a series of 11 essential hypertensives in which the blood was similarly incubated with normal kidney slices and tested for VDM activity. All of the blood samples from hypertensive patients produced pronounced VDM effects following *in vitro* aerobic incubation. A series of normotensive hospital patients on a similar diet gave essentially neutral effects in the rat test both before and after *in vitro* incubation. These observations provide another point of similarity between essential hypertension in man and experimental renal hypertension.

Hepato-renal factors in circulatory homeostasis. XIX. VEM and VDM mechanisms in nutritional cirrhosis in rats. EPHRAIM SHORR and B. W. ZWEIFACH. *Dept. of Medicine, Cornell University Medical College and The New York Hospital, New York City.* Rats on a low protein (10% casein) diet supplemented with cystine develop cirrhotic-like changes in liver and kidney (Gyorgy). The poor tolerance of these rats to surgical procedures suggested that this predisposition to shock might be related to disturbances in the renal VEM and hepatic VDM mechanisms. Cirrhotic rats prepared according to Gyorgy were studied after 120–150 days on a protein-deficient diet. Saline washes of liver invariably contained VDM. There was no impairment of the ability of the liver to form VDM anaerobically *in vitro*. The significant hepatic lesion was the failure to inactivate VDM aerobically *in vitro*. The kidneys were also found to have an impairment of their VEM mechanism, little or no VEM being produced on anaerobic incubation *in vitro*. Blood from cirrhotic rats produced in normal test rats a moderate vaso-depressor response in the terminal mesenteric arterioles and precapillaries, indicative of the presence of VDM. The mesenteric blood vessels of cirrhotic rats were hypo-responsive to the topical application of epinephrine. These phenomena are considered to reflect the derangements of the VEM-VDM mechanisms in the cirrhotic rat, as a result of which the vascular bed is chronically under the decompensatory influence of VDM. These derangements in the VDM-VEM mechanisms, because of their significance for compensatory vascular reactions to shock, are believed to contribute to the predisposition of the cirrhotic rat to shock.

The leakage of potassium from injured muscle fibres. F. J. SICHEL and CHERYL PARKHURST (by invitation). *University of Vermont College of Medicine.* Previously the rate of potassium loss from transversely cut frog skeletal muscle fibres into a restricted volume of Ringer's solution was reported. The rate was sufficient to account for conduction failure within a few minutes. Since this failure involves the entire length of each fibre in such a short time, the possibility must be considered that local trauma might change the entire surface of the fibre so that potassium loss occurs through this surface and not only through the cut ends. The surface shows no visible changes except where cut, although permeability might be altered. If the potassium came out mostly through the uncut surfaces there should be very little difference in

the rate of potassium loss in fibres cut once and cut three times. If the loss were mostly through the cut ends, there should be a greater rate of loss with more cut ends.

The muscles, in a restricted volume of Ringer's solution, were cut transversely either once or three times. The rate of accumulation of potassium in the medium was determined by means of a flame photometer. It was found that the rate of leakage of potassium was much higher in the case of those muscles with three transverse cuts than in those with one. It is concluded therefore that the loss of potassium is entirely through the cut ends or, if there is any loss through the rest of the surface this must be of a much smaller order of magnitude.

A study of insulin hypersensitivity in hypophysectomized dogs. I. H. SLATER (by invitation), R. C. DE BODO, H. F. WEISBERG (by invitation), S. P. KIANG (by invitation). *Department of Pharmacology, New York University College of Medicine.* Hypersensitivity to insulin is characteristic of hypophysectomized animals. The degree, development and possible modification of this abnormality was studied in a series of hypophysectomized dogs. The test dose of insulin (0.025 units/kgm. intravenously) in normal dogs causes either no change in blood sugar or only a slight drop of short duration (recovery within 40 minutes). Two–three days after hypophysectomy a significantly increased response to insulin was apparent, the blood sugar consistently showed a drop and the recovery was delayed. Ten–fourteen days after operation the insulin hypoglycemia was more marked and the return to the basal level greatly delayed, often beyond four hours. During the fifth postoperative week and thereafter, insulin lowered the blood sugar to near convulsive levels with only rare recovery within four hours. Since the shape of the blood sugar curves obtained after insulin differ in normal and hypophysectomized dogs, the degree of hypersensitivity cannot be expressed numerically. Whereas in normal animals 1.5 units/kg. insulin, 60 times the test dose, produces a hypoglycemia approximating the convulsive level, recovery occurs consistently within four hours. Furthermore, hypophysectomized animals have a significantly lower postabsorptive blood sugar, thus the insulin action starts at a lower level. The gradual development of the hypersensitivity suggests a relation to the secondary atrophy of the adrenal cortex and thyroid which might be preceded by a hypofunction undetectable histologically. Indeed, adrenal cortical extract modified somewhat the insulin induced hypoglycemic response.

Some cutaneous responses to "reflex cooling". DOUGLAS E. SMITH, WALTER C. RANDALL, and ALRICK B. HERTZMAN. *Department of Physiology, St. Louis University, School of Medicine.* Simultaneous estimations of skin blood flow and skin surface temperature were made upon the finger, forearm, and cheek of normal men subjected to "reflex cooling." Immediately upon exposure of the legs and lower trunk to a cold water bath, the blood flow in the finger decreased markedly but underwent little change thereafter during the exposure. The skin temperature fell markedly but at a slower rate than the blood flow and continued to fall for the rest of the period of exposure to cold. The blood flow of the forearm decreased significantly immediately upon reflex cooling and either stayed low or rose to normal levels thereafter. At the same time the forearm skin temperature gradually fell. The responses of the cheek were variable, the flow being either increased or decreased. The cheek temperatures usually followed the direction of the cheek blood flow. During reflex cooling, oral temperature was observed to rise to a peak and then fall, reaching values below the control level. The relative degrees of the changes in blood flow and skin temperature in the various areas and their possible significance are discussed.

Some toxic effects of acid and neutralized thiamine solutions. JAY A. SMITH (by invitation), PIERO P. FOA, HARRIET R. WEINSTEIN (by invitation), A. SIDNEY LUDWIG (by invitation), and J. MARVIN WERTHEIM (by invitation). *Department of Physiology and Pharmacology, The Chicago Medical School, 710 S. Wolcott Avenue, Chicago, Illinois.* Thiamine hydrochloride solutions are acid. Upon injecting such solutions intravenously, respiration is depressed, and blood pressure decreases; these effects are transitory if artificial respiration is provided. It was desired to know if these changes were due to the thiamine or to the acidity of the solution. Thiamine hydrochloride solutions weakened or stopped the isolated perfused turtle heart; neutralized solutions depressed the heart slightly; acidified Ringer depressed the heart less than a thiamine solution at identical pH. Thiamine hydrochloride solutions caused vasodilation in the perfused rabbit ear in proportion to the concentration and the acidity. Neutralized thiamine solutions caused little change in the rate of perfusion. Thiamine hydrochloride depressed the heart in the heart-lung preparation only in concentrations sufficiently great to alter the pH of the blood appreciably. A thiamine hydrochloride solution (pH 3) intravenously to dogs altered the pH of the blood only slightly (from 7.54 to 7.52) within 20 seconds. The injection of identical quantities of a neutralized thiamine solution caused no shift in the pH. The effects on respiration and blood pressure were identical after injection of acid or neutralized thiamine solutions. It is concluded that in isolated systems, such as the turtle heart, rabbit ear, and heart-lung preparation, the effects of thiamine hydrochloride are due principally, although not exclusively, to the acidity of the solutions. In intact

dogs, on the other hand, the effects of thiamine are not due to the acidity of the solutions but to thiamine itself.

Experimental embolism of selected portions of pulmonary arterial bed. JOHN R. SMITH and MASAUKI HARA (by invitation). *Department of Internal Medicine, Washington University School of Medicine, St. Louis.* Using anesthetized, heparinized dogs with chests opened, fine catheters were introduced into the main pumonay artery. The catheters could be guided into any lobar arterial branch. Minute or particulate emboli were then placed in a given lobar artery in selected quantity. Care was taken to prevent retrograde passage and scattering of emboli outside of the selected lung lobe. When small foreign bodies such as fine glass beads, poppy seeds or lycopodium spores were impacted in the lobar artery in large numbers, there were no significant effects upon cardiovascular dynamics. In contrast, the injection of very fine material, such as starch or barium sulfate suspensions, into the lobar artery produced a marked rise of pulmonary arterial pressure, a precipitous fall of systemic blood pressure, and intense distension of the right heart. Death occurred in 2 to 3 minutes. Since embolic particles such as beads and lycopodium produce no circulatory dynamic changes, and finer materials (starch or barium sulfate) produce severe circulatory changes, it is reasoned that pulmonary "capillary" emboli, affecting a restricted portion of the pulmonary vascular bed, may cause death with apparent pulmonary spasm and circulatory failure.

The influence of thiourea and thiouracil on the response of rats to methyl chloride. WILLIE W. SMITH. *Laboratory of Physical Biology, National Institute of Health, Bethesda, Maryland.* During an investigation of protective action by sulfur-containing compounds, thiourea and thiouracil were tested. Month-old rats in groups of 10 were given 20% casein diets 2 weeks preceding exposures to methyl chloride 2,000 p.p.m. 6 hours a day 6 days a week. Median survival time was about 10 days. With supplements of thiourea (0.46%) or thiouracil (0.77%), death occurred during or immediately after a single exposure. Again all rats died on one exposure with thiourea 0.05%; 8 died with thiouracil 0.1%, but only 2 with thiouracil 0.05%. With the supplements for only 3 days preceding exposure 9 on thiourea 0.06% and 8 on thiouracil 0.1% died. When thiourea (12 mg.) was injected intraperitoneally immediately before, 30 minutes after, or 2 hours after the exposure, all of the rats died. Of those injected 30 minutes after, 7 died within 2 hours evidencing extreme respiratory distress. Only 2 of the rats injected 18 hours after exposure died. With supplementary cystine, 0.45%, rats survive about 8 weeks. Thiourea 0.06% in this diet resulted in 100% mortality on a single exposure; with thiouracil 0.1% 6 died within 2 days, 3 lived longer than 8 weeks. 6-Methyl-2-thiouracil, 6-propyl-2-thiouracil and sulfadiazine were fed for 3 days in molar concentration 5 times that of 0.1% thiouracil. Half of the rats on methylthiouracil died after 1 or 2 exposures, but the other compounds had little or no effect. The action of thiourea and thiouracil on methyl chloride-exposed rats does not parallel their known effect on thyroid action.

Excretion of bilirubin and bromsulfalein by the liver. W. J. SN[illegible]PE (by invitation), C. W. WIRTS (by invitation), [illegible]. L. MILLER and A. CANTAROW. *Jefferson Medical College, Philadelphia, Pa.* Employing Thomas-type bile-fistula dogs, bile was collected continuously, in 15-minute samples, before, during and after intravenous injection of bilirubin and of bromsulfalein. The duration of injection and post-injection periods was varied in different experiments. Serial determinations were made of the concentrations of bilirubin and bromsulfalein in the serum and of the volume, and total bile pigment and dye concentrations in the bile in 15-minute samples. After a single injection of 1 mg. bilirubin per kilogram, 48.9–89% of the amount injected was recovered in the bile in 3 hours. After a single injection of 5 mg. bromsulfalein per kilogram, 55.6–96.9% was recovered in the bile in 3 hours. After continuous injection (½–2 hours) of 0.7–5 mg. bilirubin per kilogram, 61–100% of the amount injected was recovered in the bile in 2–4 hours. After continuous injection of 4.5–12 mg. bromsulfalein per kilogram (2 hours), 48–74.7% was recovered in the bile in 4 hours. The data indicate that injected bilirubin and bromsulfalein are removed from the blood more rapidly than they are excreted in the bile. If the liver is the only organ involved in their removal from the blood, this implies a subsequent phase of temporary storage in that organ prior to passage into the bile.

Facilitation produced by cerebellar stimulation. RAY S. SNIDER (by invitation) and H. W. MAGOUN. *Department of Anatomy, Northwestern University, School of Medicine, Chicago, Illinois.* In monkeys excitation of (a) anterior lobe-lobulus simplex region and (b) paramedian lobule-tuber vermis-pyramis region of the cerebellum produced facilitation of reflex activity and of movements evoked from the cerebral cortex. The most pronounced effects were noted when chloralosane anesthesia was used and electrical stimuli of 200–300 cycles per second were applied to the cerebellum from 5–15 seconds before the elicitation of the movement. Facilitation occurred following destruction of the superior peduncle suggesting that the cerebellum is exerting its influence through facilitatory centers in the brain stem rather than through the activation of cerebral mechanisms. The distribution of cerebellar facilitatory areas in the monkey is similar to that of the tactile receiving areas and of the

areas yielding inhibition of movement in the cat. This similarity even extends to the somatotopic representation of body parts for, though overlapping was present, within zone (a) movements of the leg were facilitated from points rostral to those for the arm, while within zone (b) the reverse was the case.

Oxygenation of the cord blood of breathing fetuses during a prolonged period. FRANKLIN F. SNYDER. *Harvard Medical School.* The persistence of rhythmical respiratory movements in rabbit fetuses following their exposure in a saline bath with intact umbilical cord circulation, affords opportunity to sample the umbilical vein blood of breathing fetuses at various intervals after their escape from the uterus. The state of activity of the fetus and the degree of sensitivity to tactile stimuli can be correlated with the oxygen supply. Following the inhibition of labor by the induction of ovulation and a fresh growth of lutein tissue near term, the uterus is quiescent and the placenta remains attached even in fullterm and postmature rabbits. In fullterm fetuses under continuous observation for an hour or longer while showing rhythmical respiratory movements it is evident that the blood of the umbilical vein may remain bright red in contrast to the blue blood of the arteries. Determination of the oxygen content in 28 blood samples obtained from the umbilical veins of 13 fullterm fetuses showed that at the beginning and at the end of one hour, the oxygen content averaged 11 volumes per cent; the percentage saturation with oxygen averaged 73 at the beginning and 78 at the end of one hour. The carbon dioxide content of the blood was lower at the end of an hour of exposure in the saline bath than at the beginning.

Pulmonary function as affected by operative positions. A. SOKALCHUK (by invitation), D. ELLIS (by invitation) and E. M. GREISHEIMER. *Department of Physiology, Temple University School of Medicine.* We have determined rate, tidal volume, oxygen use, maximal breathing capacity and vital capacity, and calculated from these minute volume of respiration, ventilatory equivalent and per cent reserve. The twelve positions studied are those used in the operating room of Temple University Hospital. Each subject has been observed three times in each position. The supine position is used as the control, and the results in the other positions are compared with this. The following trends have been noted in the subjects studied to date. The rate is faster in the sitting and jack-knife positions. The tidal volume is lower in the sitting, both lateral and both kidney positions. It is higher in the Trendelenburg position. The oxygen use is greater in the sitting, prone and lithotomy positions. The maximal breathing capacity is greater in the sitting position. It is decreased in the Trendelenburg, both lateral, both kidney and gall bladder positions. The vital capacity is greater in the sitting position. It is lower in the prone, Trendelenburg, right lateral, both kidney, gall bladder, lithotomy and jack-knife positions. The minute volume of respiration is decreased in the reverse Trendelenburg, both kidney and gall bladder positions. The ventilatory equivalent is lower in the sitting, Trendelenburg, left lateral, both kidney and lithotomy positions. The per cent reserve is lower only in the lithotomy position.

The mechanism of nitrous oxide analgesia. R. R. SONNENSCHEIN, R. JAMISON, L. LOVSETH, W. CASSELS (by invitation) and A. C. IVY. *Depts. of Clinical Science and Anesthesia, University of Illinois College of Medicine, Chicago.* The pain threshold-raising action of nitrous oxide, in concentrations of 10% to 40% in oxygen, was studied by the method of electrical stimulation of the tooth pulp (Goetzl, F. R., Burrill, D. Y., and Ivy, A. C.: Quart. Bull., Northwestern U. Med. Sch., **17**: 280, 1943). Control and test determinations were made at one to two minute intervals. Psychomotor activity was tested before and during gas administration, by means of the Johnson Code Test.

Forty-one tests on eighteen subjects revealed a significant rise in threshold of 0.30 to 0.45 volts (approximately 25%), at concentrations of 20%, 33%, and 40% N_2O; results at 10% were variable. Results of twenty-two psychomotor tests during gas administration showed an increase of 14% to 40% in the standard test time at concentrations of 10%, 20%, 33%, and 40%. Readings on four occasions with the Millikan oxymeter, at 33% N_2O, showed no change in oxygen saturation, over the control readings.

It is concluded that the analgesic action of nitrous oxide, as a typical general anesthetic agent, is associated with a diminution in psychomotor activity. It appears likely that the analgesic action may be secondary to the latter factor.

Performance in warm environments following hemorrhage, albumin infusion, bed rest and exposure to cold. C. R. SPEALMAN, E. W. BIXBY (by invitation), J. LARUE WILEY (by invitation), MICHAEL NEWTON (by invitation) and H. C. BAZETT. *Department of Physiology, University of Pennsylvania, Philadelphia.* Deterioration of ability in certain physical activities (endurance and tilt table tests) can be detected at normal temperatures following the removal by venesection of 500 to 1000 cc. of blood (Karpovitch and Millman, Res. Quart. **13**: 166, 1942; Green and Metheny, Surg. Gyn. Obs. **84**: 1045, 1947). One would expect even more pronounced effects with unacclimatized individuals in very warm environments which necessitate the physiologic stress of maintaining blood volume adequate for a vascular system enlarged by dilatation of blood vessels in the skin. In our experiments, performance (tilt table, bi-

cycle ergometer, Crampton test) in the heat (33°C., D. B., 28-29°C., W. B., wind velocity 50 cm./sec.) became markedly poorer shortly after the withdrawal of 500 cc. of blood and remained so for several days. Removal of 200 cc. of blood also resulted in decreased ability to perform. Evidence that the causal factor was a decrease in blood volume rather than a decrease in hemoglobin was obtained in experiments in which performance was similarly affected by decreasing plasma volume alone (induced by bed rest and exposure to cold). Furthermore, increasing blood volume by infusing serum albumin (in an amount equivalent to 500 cc. of plasma) improved performance in the heat. Eight normal healthy young men served as subjects for these experiments.

Electroencephalographic studies following thalamic lesions in humans. E. A. SPIEGEL and H. T. WYCIS (by invitation), *Dept. of Experimental Neurology, Temple Univ. School of Medicine, Phila., Pa.* Circumscribed lesions were placed by electrocoagulation in the region of the medial nuclei of the thalamus (medial thalamotomy) for treatment of some mental disorders associated with emotional tension; or thalamotomy was combined with lesion of the spinothalamic system in the midbrain in cases of intractable pain. The stereotaxic technique previously described by us (Science **106:** 349, 1947) was used for introducing the coagulating electrode into the brain. The electroencephalogram was recorded from the scalp before and after operation. In some instances also a basal electrode was applied through the nose so that it touched the posterior wall of the nasopharynx. A comparison of the electrograms with those obtained after prefrontal lobotomy showed that the disturbance of cortical function as far as revealed by the electrical discharges is much smaller after thalamotomy than after prefrontal lobotomy. However in some instances, at least a transitory slowing of the brain waves, particularly in the frontal leads, could be demonstrated, apparently due to loss or diminution of the impulses from the medial nuclei to the frontal poles. The results are compared with similar studies on monkeys.

Protection by substrate against inhibition of enzymatic adaptation by protein denaturants. S. SPIEGELMAN and JOHN M. REINER *Department of Bacteriology and Immunology, Washington University School of Medicine, St. Louis, Mo.* Previous studies (Arch. Biochem., **13:** 113, 1947) have indicated that enzymatic adaptation involves a modification in the protein components of the cell. One of the problems raised by this phenomenon is the nature of the precursor which is converted into active enzyme by the presence of substrate. In particular, the question arises as to how close a relation exists between the precursor and the final active enzyme. A method of throwing some light on this problem was provided by the finding that protein denaturants, such as urea, guanidine and desoxycholate exerted strong inhibitory effects on the adaptive process. These experiments indicated that a denaturable molecule was an important component of the adaptive system. If this component was sufficiently similar to the active enzyme so that it could bind with substrate, it would be expected that the presence of substrate would protect it from inactivation. Experiments performed to test this possibility confirmed the expectation. Unadapted cells treated with any of these agents previous to incubation with substrate took two-three times as long to show evidences of enzyme formation. If, however, the substrate was added first and then the denaturing agent, no inhibition of enzyme formation was observed. These experiments would therefore indicate that a component, present in unadapted cells and formed during enzymatic adaptation, although not enzymatically active, can combine with substrate. The combination with substrate protects the component against inactivation by protein denaturants.

Studies on diffusion respiration. v. The hemoglobin-oxygen pump. JOSEPH N. SPENCER (by invitation), WILLIAM B. DRAPER, THOMAS M. PARRY (by invitation) and RICHARD W. WHITEHEAD. *From the Department of Physiology and Pharmacology, University of Colorado Medical Center.* Draper and Whitehead (1944) reported that animals placed in respiratory arrest in an atmosphere of oxygen may survive for considerable periods. They attributed their results to the continuous vis a fronte exerted on the atmosphere of the respiratory spaces by the hemoglobin-oxygen pump. The functional importance of this pump is illustrated by the following experiments. Dogs, following denitrogenation, were connected with an oxygen filled, sensitive spirometer, placed in respiratory arrest with thiopental or curare and the oxygen uptake determined. In five experiments lasting forty-five minutes or longer, there was essentially no difference in oxygen uptake between the animals receiving thiopental and those receiving curare. With respiratory arrest, oxygen uptake decreased approximately 11 per cent. This level was maintained for thirty minutes, then gradually fell to reach a level 30% less than the control during the last few minutes of life, ceasing abruptly with cardiac arrest. In a second series of five experiments the spirometer bell was seized following respiratory arrest. The maximum negative intrapneumonic pressure developed in the series exceeded 10 cm. of water. In all instances where the spirometer bell was seized, death was associated with a profound collapse of the lungs. In a third series of experiments the animals, after respiratory arrest, were connected to a spirometer filled with

a carbon monoxide-oxygen mixture. The atmosphere was moved en masse and carbon monoxide appeared in the blood in proportion to its concentration in the spirometer.

A mass spectrometer for the rapid, continuous analysis of respiratory gases. By RALPH W. STACY, (by invitation), JACK A. HUNTER, (by invitation), and F. A. HITCHCOCK. *The Laboratory of Aviation Physiology, The Ohio State University, Columbus.* To make possible a satisfactory study of the changes in composition of alveolar air that occur following rapidly induced changes in ambient pressure it was necessary to develop an analytical instrument that would record accurately rapid changes in composition of the air leaving the lungs. We have, therefore, designed and constructed a mass spectrometer of the Neir type, specially adapted for the measurement of respiratory gases. The instrument makes possible the simultaneous analysis for oxygen, nitrogen, and carbon dioxide. A special calibrating arrangement permits the quantitation of each of these components. The instrument records the concentrations of all three components on a single photographic film. The ion source is so constructed that analysis is continuous. The time required for recording full scale concentration change is less than two tenths of a second. The analytical error of the instrument is less than two per cent of full scale in normal use. An expanded scale provision has been incorporated which magnifies the changes, thus increasing the accuracy. Preliminary analyses of alveolar air expulsions have not verified the existence of an oxygen trough. This problem is being studied further.

Effect of anoxia on the glucose tolerance curve of dogs. J. CLIFFORD STICKNEY, DAVID W. NORTHUP and EDWARD J. VAN LIERE. *Dept. Physiology, School of Medicine, West Virginia Univ., Morgantown.* Seven fasted, unanesthetized dogs were given intravenously 1.5 g. glucose per kilogram body weight. Blood sugar concentrations were determined by the Folin Wu method on samples drawn immediately before and at 15, 30, 60 and 90 minutes after the injection. Glucose tolerance curves were likewise determined while the dogs were subjected (except for brief sampling periods) to a barometric pressure of 254 mm. Hg. The hyperglycemic response of the dogs to the same barometric pressure was also determined on samples drawn at the above time intervals. In most cases the response of each dog in the three types of experiment was determined three times, but at weekly intervals to diminish the cumulative effects of repeated glucose injections as well as of repeated exposures to anoxia. Fifteen minutes following glucose injection, the blood sugar values averaged 266 per cent above the control. At 60 min. they had fallen uniformly to 29 per cent above, and at 90 min. were at or below the control. Anoxia tended to reduce the height of the 15-minute maximum of the glucose tolerance curve, but slowed the rate of fall beyond that point so that the 90 min. value was still 33 per cent above the control. Anoxia alone, caused a 32 per cent rise in blood sugar which fell off gradually to 19 per cent at 90 min.

Direct experimental comparison of several surface temperature measuring devices. ALICE M. STOLL (by invitation) and JAMES D. HARDY. *From the Russell Sage Institute of Pathology, The New York Hospital, and the Department of Physiology, Cornell University Medical College, New York, N. Y.* This paper presents comparative data on several surface temperature measuring devices used in a variety of experimental conditions simulating natural climatic environments. The instruments studied included the radiometer, a resistance wire thermometer, surface pyrometers, several different types of wire thermocouples and mercury in glass thermometers. An apparatus for providing a standard surface whose temperature could be determined within $\pm$ 0.05°C. under all experimental conditions is described. The simulated environments employed were: normal room conditions, forced convection, infrared radiation, "sunlight" radiation, and "sunlight" plus forced convection. The results of this study indicate that the radiometer is the most dependable of the instruments studied. With this instrument an accuracy of $\pm$0.1°C. was maintained under all conditions whereas measurements made with other instruments of equal accuracy under room conditions were affected seriously by radiation and forced convection. The advantages and disadvantages in the application of each instrument to the accurate measure of skin temperature are discussed on the basis of the experimental findings.

Olfactory acuity and appetite: effects of bitter tonics on olfactory acuity in normal human subjects. FREYA STONE (by invitation) and FRANZ R. GOETZL. *Department of Medical Research, Permanente Foundation, Oakland, California.* Olfactory thresholds (Elsberg's method) were determined in normal individuals daily, at regular intervals. On test days the subjects' statements regarding appetite and their caloric intake were recorded. Meals were found to be preceded by a period of increasing and followed by one of increasing olfactory acuity. The precibal increase in olfactory acuity could be prevented by intercibal ingestion of food, the decrease in that acuity by omission of meals. It is suggested that by estimating diurnal variations in olfactory acuity measures may be found for the sensations of appetite and satiety. Benzedrine was observed to produce decrease in olfactory acuity, decrease in the intensity of the sensation of appetite, decrease in caloric

value of freely selected meals and, also, a sensation of satiety. Bitter tonics ingested between meals failed to influence precibal increase in olfactory acuity, intensity of the sensation of appetite, and caloric values of succeeding meals. The absence of effects of bitter tonics may be explained by assuming refractoriness to augmentation in normal individuals of precibal increase in olfactory acuity and intensity of the sensation of appetite, both presumably of optimal magnitude. When ingested during meals, however, bitter tonics were found to prevent the postcibal decrease in olfactory acuity and to render incapable freely selected meals of bringing about the conversion of the sensation of appetite into one of satiety.

The question whether the effects of bitter tonics described are related to appetite stimulating effectiveness commonly ascribed to bitter agents is being subjected to further investigation.

Adjustment of caloric intake of rats to dietary change. JACK L. STROMINGER (by invitation) and JOHN R. BROBECK. *Laby. of Physiology, Yale Univ. School of Medicine, New Haven, Conn.* When 43% fat was added to the calf meal diet of 222 gm. male albino rats, during the first five days they ate 132% of the calories taken by controls. During the next 19 days they ate the same number of calories as controls. When 43% carbohydrate was added to the calf meal diet of similar rats, during the first five days they ate 91% of the calories of controls. During the next 19 days they ate the same number of calories as controls. When 30% or 60% fat was added to the calf meal diet of 101 gm. male albino rats, during the first 5 days they ate 117% of the calories of controls. For the next 17 days they ate 148% of the calories of controls. When 30% or 60% carbohydrate was added to the calf meal diet of similar animals, during the 22-day period they ate the same number of calories as controls. When 50% fat was added to the calf meal diet of obese female rats with hypothalamic hyperphagia, for 21 days they ate about 200% of the calories they had taken on the calf meal diet. Female unoperated controls ate about 200% for the first day, and thereafter diminished their caloric intake until by 5–7 days they were eating the same number of calories they had taken on the calf meal diet. The operated animals showed marked acceleration of weight gain during this period, while the controls gained at a normal or slightly diminished rate.

The recovery period after resuscitation. H. G. SWANN and MARSHALL BRUCER (by invitation). *Dept. of Physiology, University of Texas Medical School, Galveston.* Dogs were subjected to the anoxia of breathing pure nitrogen or 2.43% oxygen in nitrogen or 1% carbon monoxide in air. When death was imminent, various measures to ventilate the lungs were instituted (see M. Brucer and H. G. Swann, this journal) in order to resuscitate the animals. The first sign of recovery was a sharp rise in systolic blood pressure, but not necessarily a rise in diastolic pressure. Soon thereafter breathing was reinstated. The first rise in blood pressure occurred within 100 seconds or not at all, even though the pulmonary ventilation was continued for 20 minutes. If good heart action was not resumed within 100 seconds, then the attempted resuscitation failed and further pulmonary ventilation was futile. When cardiac massage as well as artificial respiration was instituted, recovery from lower levels was accomplished. However the return of blood pressure and respiration was, slower. In all but one instance, the animals showed various degrees of dementia,—i. e. they were blind or comatose or exhibited signs of decerebrate rigidity. This dementia, presumably due to the anoxic experience, was also frequently observed in carbon monoxide—poisoned dogs which were resuscitated only by measures ventilating the lungs.

Principles of protection against the effects of negative "G". (Motion Picture). HENRY M. SWEENEY and Biophysics Branch personnel. *Aero Medical Laby., Air Materiel Command, Wright Field, Dayton, Ohio.* A colored motion picture was made to show methods of protecting against the effects of negative acceleration. The development of hemorrhage and petechiae in the sinuses and conjunctivae is shown in man and animals. Motion pictures of the blood vessels of the brain of a monkey with a lucite calvarium under g show that within the closed box of the skull fluid counterpressure protects against blood vessel rupture. The technique of radial arterial and frontal vein canullation using an Ungar type double needle is demonstrated together with the use of a Gauer-Wetterer inductance pressure gauge for measurement of venous and arterial-pressure in man while undergoing negative acceleration. This film shows pictures of humans undergoing abrupt accelerations demonstrating the greater tolerance achieved by shortening the period of application of g. The principle of protection afforded by inclination of the long axis of the body to the direction of the g-force is demonstrated.

The effect of adrenal cortical extract on adrenal response to total body X-irradiation. MARGUERITE N. SWIFT, HARVEY M. PATT and ELLA B. TYREE (introduced by AUSTIN M. BRUES). *Biology Division, Argonne National Laboratory, Chicago, Illinois.* The transient 20–50% decrease in adrenal total cholesterol observed in albino male rats 3 hours after a midlethal total body exposure to X-radiation (Patt et al. Am. J. Physiol. **150**: 480, 1947) is largely prevented by subcutaneous injection of adrenal cortical extract (Wilson). Total doses of 0.3–1.8 cc./200 gm. rat were similar in effect, with no graded response over this range. This is additional evidence that the early choles-

terol fall reflects increased adrenal activity. In order to investigate the nature of the marked rise (70–109%) in adrenal cholesterol observed 3–7 days after a midlethal exposure to X-radiation, a second similar exposure was administered to rats 4 days after the first. No fall in adrenal cholesterol occurred by 3 hours after the second exposure, and the cholesterol level at this time was unaffected by cortical extract dosage sufficient to prevent the early decrease seen after a single X-ray exposure. Elevated adrenal cholesterol is reported to occur only after prolonged stimulation by adrenotrophic hormone, and may in this case also represent overstimulation in excess of cortical hormone demand. Rats given 0.1 cc. of extract three times daily, an amount sufficient to prevent the early cholesterol fall, show adrenal cholesterol changes of slightly lesser degree than do controls over a 15 day post-irradiation period. Survival is not enhanced by this treatment. Larger amounts of extract might possibly prove effective since increased survival has been reported after X-irradiation by shielding the adrenals (Craver, Fed. Proc. **6:** 319, 1947).

Silicone implantations in the rat. VERNE W. SWIGERT (by invitation), HAMILTON R. FISHBACK (by invitation), LILLIAN E. CISLER (by invitation), and FREDERIC T. JUNG. *Northwestern University Medical School, Chicago 11, Ill.* Weighed samples of two types of silicone were implanted aseptically into the abdominal cavities of white rats. The first, a solid type of silicone, similar in appearance to white rubber, was in the form of cylindrical pieces, about 6 mm. in diameter. The second type was a variety called "bouncing putty" because it flows like an opaque grayish liquid with low surface tension, but slowly, and bounces if dropped on the floor. The bouncing putty was used in a series of 61 operations beginning 1946 Jan. 28; the solid type was used in only 4 rats beginning 1946 Feb. 18. The animals were sacrificed for necropsy at times varying from 2 months to a year. There were no indications in the behavior of the animals, or in their weight curves, or in the necropsy findings, that absorption or any chemical change in the silicones had occurred during that time. The bouncing putty remained recognizable as grayish deposits of varying size, sometimes very finely divided and often widespread, on both parietal and visceral aspects of the peritoneum. The silicone was not transported to deeper or remoter structures, but remained on the surface. It was enveloped in a thin layerof connective tissue that contained occasional giant cells and showed signs of mild chronic reaction. One rat developed a tumor that grew rapidly until it constituted 29% of the body weight; since the tumor was extra-abdominal, its significance was debatable.

The chemical composition of regenerating rat liver: the influence of ovariectomy. CLARA M. SZEGO and SIDNEY ROBERTS. *Department of Physiological Chemistry, Yale University, New Haven.* In the adult female rat, the liver stump remaining after removal of two-thirds of the liver doubled its weight within two days. During the first day, the percentage of fat in the liver reached a maximum of 12% (normal, 6%) and the water content dropped to 67% (normal, 71%). The change in per cent protein was apparently relatively small. On a body weight basis, total liver lipid returned to normal within 2 days (250 to 300 mgm. per 100 gm.). Subsequently, as liver restoration progressed, the percentage of water and fat approached normal levels. Rats ovariectomized two weeks prior to partial hepatectomy exhibited overall changes similar to those observed in non-ovariectomized animals. A profound difference was, however, noted during the first 8 to 10 hours following operation. In control animals, hepatic fat mobilization occurred essentially at a constant rate for the entire first day (ca. 8 mgm. per 100 gm. body wt. per hr.); a concomitant decrease in liver water was observed. On the other hand, in the ovariectomized group there was an initial lag (first 10 hours) in fat deposition, accompanied by a rapid uptake of water (70.6% to 72.5%). Thereafter, the rate of fat mobilization was similar in both groups, so that the maximum lipid concentration reached in the ovariectomized group was only about 10%. Preliminary observations indicated that the administration of estrogen to the ovariectomized rat immediately after partial hepatectomy may restore the initial rate of fat transport to the liver to normal.

Effects of the cold pressor test on glomerular filtration and effective renal plasma flow. P. J. TALSO (by invitation), A. P. CROSLEY, JR. (by invitation), and R. W. CLARKE. *Medical Department Field Research Laboratory, Fort Knox, Ky.* Studies were made of the effects of the cold pressor test on renal function. Seven male volunteers who had no history of renal disease served as subjects. Glomerular filtration (as measured by mannitol clearance) and effective renal plasma flow (as measured by sodium para-aminohippurate clearance) were determined before, during and after immersion of the foot in ice water at 1°C. for 15 minutes. In 6 of 7 subjects both glomerular filtration rate and effective renal plasma flow decreased either during the application of the cold stimulus or within approximately 30 minutes thereafter. In no subject did the effect persist longer. The average decreases in glomerular filtration rate and effective renal plasma flow, as compared with the control values, were 14% and 21%, respectively.

The behavior of certain characteristics related to exhausting work during recovery from semi-starvation. HENRY LONGSTREET TAYLOR, AUSTIN HENSCHEL, JOSEF BROZEK, and ANCEL KEYS. *Laboratory of Physiological Hygiene, University*

of Minnesota, Minneapolis. Six men who lost 24 per cent of their body weight during a six months regimen on a famine diet characteristic of Western Europe were studied during the 20 weeks of refeeding necessary to regain the original body weight. Maximal oxygen intake, 12-minute recovery lactate and 10-minute oxygen debt were studied during and after a 3-minute run at 7 miles per hour and at a grade which depended on the capacity of the subject. The Harvard Fitness Test was used and measurements of strength were made with the hand and back dynamometer. Loss of function from 20 to 70 per cent of the control value was noted in these measurements after semi-starvation. Recovery was slow. After 12 weeks, the Harvard Fitness Test score had recovered 58 per cent and the maximal oxygen intake 42 per cent of the semi-starvation loss. After 20 weeks of refeeding, when the men had regained their weight, the maximal oxygen intake, the 12-minute lactate and the 10-minute oxygen debt had regained their normal values. The time and score of the Harvard Fitness Test recovered only 78.0 and 72.0 per cent of their respective losses, while the hand and back dynamometers had regained only 46 and 86 per cent of the strength lost. It is suggested that the poor return of strength was in a large part responsible for the failure of endurance, as measured by the Harvard Fitness Test, to recover as rapidly as the body weight.

Effects of denervation on experimental renal hypertension. ROBERT D. TAYLOR, A. C. CORCORAN and IRVINE H. PAGE. *From the Research Division of the Cleveland Clinic Foundation, Cleveland, Ohio.* High caudal or spinal anesthesia at D5 in patients suffering from essential hypertension commonly results in a decrease of arterial pressure to normal levels and an increase of minimal renal plasma flow to average 1.2 times normal. It follows that one effect of such anesthesia is a considerable decrease in renal vascular resistance. In attempting to define this phenomenon further observations were made in dogs with experimental renal hypertension due to perinephritis. Of 11 dogs given spinal or epidural anesthesia to levels causing paralysis of the nictitating membrane, arterial pressure was decreased in all (means from 172 control to 122 mm. Hg. during anesthesia) and minimal renal plasma flow increased on the average by 10 per cent in 9. Renal plasma flow was decreased in 2 dogs in which control levels were already very low. In another 2 dogs the effects of anesthesia were maintained for 36 hours. Thus, in dogs, as in patients, spinal or epidural anesthesia usually induces renal vasodilation. That this vasodilation was not due to the systemic effects of the anesthetics used (procaine and metycaine) was shown in 4 experiments in which these drugs were injected into lumbar muscles and 2 in which they were given intravenously. The effect is therefore due to the denervation caused by local drug action. Arterial pressure and renal plasma flow were determined in 2 dogs after spinal section by ligature and caudal pithing. Section at D5 did not alter these functions but section at C6 or C8 had the qualitative effects of high spinal or epidural anesthesia. Similarly, autonomic denervation by intravenous infusion of 2 hypertensive dogs with ET_4N decreased arterial pressure while renal plasma flow was unchanged or slightly increased.

It is concluded that maintenance of experimental renal, as of clinical arterial hypertension, is partially dependent on an unidentified vasomotor outflow from the upper thoracic spinal cord.

Response of deafferented neurones of spinal cord to impulses reaching them from higher levels of central nervous system. R. D. TEASDALL (by invitation) and G. W. STAVRAKY. *Department of Physiology, Faculty of Medicine, University of Western Ontario, London, Canada.* Decentralization is known to increase the sensitivity of spinal neurones to chemical stimulating agents (Cannon, Am. J. Med. Sci. **198**, 737, 1939. Stavraky, Am. J. Physiol. **150**, 37, 1947). It was shown recently that deafferentation leads to similar results (Stavraky and Drake, Federat. Proc. **6**, 211, 1947). The latter finding provided a method by which to determine whether the increased irritability of partially denervated neurones is limited to a greater effectiveness of chemical agents upon such neurones, or whether they will respond more readily to nerve impulses reaching them along pyramidal tracts and other descending systems of nerve fibres as well. In cats, 10 days to 3 months after aseptic deafferentation of the hind limb, decerebration and electric stimulation of the region of the cerebral peduncles on the transected surface of the brain was carried out. The threshold of the response to faradic stimulation of the corresponding cerebral peduncle was markedly lowered by preceding deafferentation of the limb. Furthermore, the movements observed in the deafferented limb on stimulation of the cerebral peduncle were sustained after the end of stimulation over longer periods of time than movements of the intact limb. A study of the labyrinthine and neck reflexes in deafferented fore and hind limbs of chronic animals is in progress and a demonstrable hyperactivity of reflex mechanisms is found to be present. All these results indicate that partial denervation increases the irritability of the neurones in the central nervous system, not only to chemical stimulating agents, but to physiological nerve impulses traversing intact synapses.

Critical flicker frequency in defective fields of vision. HANS-LUKAS TEUBER (by invitation) and MORRIS B. BENDER. *From the Psychophysiological Laboratory, Department of Neurology, New York*

University, College of Medicine. In 28 Naval casualties with penetrating wounds of the occiput, critical frequencies for fusion of visual flicker (c.f.f.) were significantly below those of 20 normal controls. This was found in the center of the visual fields and at increasingly eccentric points in each quadrant. In the defective fields, the per cent loss in c.f.f. (as compared with corresponding normal values) was minimal in a zone 10° from the fixation point. With increasing eccentricity the loss in c.f.f. increased. Maximal reduction of c.f.f. occurred in homonymous half fields or quadrants. In the same areas of greatest depression of c.f.f. there were maximal disturbances in perception of apparent (stroboscopic) and actual movement; the apparent speed of objects moving through these areas of maximum depression of c.f.f. was increased. This was demonstrated by moving targets at variable rates through different areas of the field, and by having the subjects manipulate the target speeds until the speeds appeared subjectively equal. Thus objects moving at various standard speeds appeared to the subjects with trauma, but not to the controls, as either abnormally fast, elongated or multiplied. It was concluded: 1) neural rather than photochemical factors limit flicker perception in these brain injured subjects; 2) the limiting factor is an (abnormally slow) intermittence of cerebral function subserving vision; 3) one and the same functional mechanism determines perception of flicker, apparent motion and actual motion.

Comparison of mannitol and thiocyanate volumes in several patho-physiological conditions. C. P. THARP (introduced by R. R. OVERMAN). *Department of Physiology, University of Tennessee, College of Medicine, Memphis, Tennessee.* Recent reports have indicated that SCN fails to remain primarily an extracellular anion in malaria and certain other diseases (J. Lab. Clin. Med. **31:** 1170, 1946). We have felt obliged, therefore, to compare the volume of distribution of mannitol, a substance of molecular size, with that of SCN in several patho-physiological conditions. Simultaneous measurements of these volumes were consequently made with the following average findings expressed as cc./kgm. body weight: (1) Normal pregnant human patients; mannitol 297 cc., SCN 282 cc. (2) Pregnant patients with eclampsia; mannitol 296 cc., SCN 281 cc. (3) Normal dogs; mannitol 287 cc., SCN 293 cc. (4) Dogs severely dehydrated by water and food deprivation for approximately 20 days; mannitol 366 cc., SCN 356 cc. It is evident from these data that mannitol and thiocyanate are diluted by approximately the same fluid volume in the normal condition, and that they continue to occupy approximately the same space in certain severe patho-physiological alterations.

In (1) the post-pyrexic state after several daily artificial temperature elevations and (2) following unilateral adrenalectomy, however, mannitol and SCN are not diluted by the same proportion of body water of the dog. In these conditions the SCN space may become as much as 64 per cent greater than the mannitol space, which remains relatively constant. It may be inferred, therefore, that while mannitol continues to be diluted only by extracellular water, SCN may enter tissue cells and be diluted by an increasing proportion of intracellular water in these conditions.

Injury potential in potassium depleted frog muscle. JULIAN M. TOBIAS. *University of Chicago Department of Physiology and Toxicity Laboratory.* In an attempt to further evaluate the assumed role of potassium in the genesis of certain bio-potentials, the injury potential has been studied, mainly by the crush technique in frog sartorii after soaking in potassium free solutions to remove potassium. Muscles soaked in normal Ringer tend to show a higher injury potential than those soaked in potassium free Ringer. In muscles soaked in normal Ringer there is a tendency, though slight, for the E.M.F. to parallel the potassium content. In muscles soaked in potassium free Ringer there is no correlation between E.M.F. and total potassium. Muscles soaked in distilled water for 5–48 hours become potassium free and lose most of their sodium. Interestingly, sodium seems the more tenaciously held. Such muscles, though potassium free, may exhibit potentials up to 30 mv. though most are lower, 9–15 mv. This potential rapidly falls to zero when outside potassium is increased to 0.1 N on the uncrushed surface of the muscle. The usual explanation that such a potential fall is due to levelling an intracellular-extracellular potassium gradient cannot apply since there is no potassium in the muscle to begin with. A few experiments with the micro-capillary electrode of Gerard et al. (with the generous help of Mr. Gilbert Ling) show the potential of the distilled water soaked muscle, truly measured between the cell interior and exterior, may be reversed from the usual orientation to as high as 40 mv. although the same muscle shows a 10–15 mv. E.M.F. of the usual orientation by the crush technique.

Sodium and potassium in insects; larvae, pupae and adults. JULIAN M. TOBIAS. *University of Chicago Department of Physiology and Toxicity Laboratory.* The high haemolymph potassium and low sodium which have been reported for certain insects have important implications for the functioning of irritable tissues, genesis of membrane potentials and the mechanisms of potassium accumulation and relative sodium exclusion by cells. In roach (Periplaneta americana) and grasshopper (Romalea microptera) haemolymph respectively, sodium (107 and 64 mM) exceeds potassium (17.3 and 18 mM), but the Na:K ratio is small (6.2 and

3.6) compared to that in the frog (41.6) and man (29.2). This is primarily because of the high potassium in the insects. In the silkworm (Bombyx mori) larva with a still higher potassium (40 mM) and very low sodium (14 mM) the ratio is less than unity (0.35). Larval haemolymph sodium (14 mM) in the silkworm may be in simple diffusion equilibrium with the sodium of the food eaten (mulberry leaf) (18 mM). Most interestingly, the entire silkworm pupa seems to contain no sodium at all or vanishingly small quantities of it. This is not true of Cecropia or Cynthia pupae. Data for the roach, grasshopper and silkworm suggest that the low sodium and high potassium are coincidental with the vegetarian food habit but not immediately causally related to it. Roach nerve and muscle can apparently function normally in a body fluid whose potassium concentration is as high as 49.2 mM. The evidence shows that tolerance for such high potassium levels need not be on a basis of potassium binding as has been postulated in the past.

Lipid inhibitors and accelerators of blood coagulation in extracts of human brain. L. M. Tocantins, R. T. Carroll (by invitation) and Thomas McBride (by invitation). *Division of Hematology, Dept. of Medicine, Jefferson Medical College, Philadelphia, Pa.* Concentrated aqueous extracts of human brain and aqueous suspensions of ether extracted lipids of the same organ contain mixtures of inhibitors and accelerators of blood coagulation. A potent lipid inhibitor, soluble in acetone, ether, alcohol, chloroform and carbon tetrachloride, heat labile (destroyed after 10 minutes at 70°C.) may be separated from the clot accelerating cephalin fraction; when freed of it, the potency of the inhibitor becomes greatly magnified. The inhibitor exerts its action on the first phase of coagulation (availability and utilization of thromboplastin); it delays the activation of prothrombin by strong thromboplastin and cephalin preparations, and enhances the antithromboplastic (and anticephalin) activities of normal plasma. The inhibitor is not heparin, is not inactivated by protamine, has no antithrombin or antifibrinogen action and differs in its behavior to heat, its solubility and method of extraction from a lipid inhibitor previously isolated (Chargaff 1937) from the spleen and other organs.

Some effects of anticholinesterases upon frog sciatic nerve. James E. P. Toman. *Dept. of Physiology, University of Utah School of Medicine, Salt Lake City, Utah.* Confiming the observations of Lorente de Nó (*Studies Rockefeller Inst. Med. Research* **131:** 195–243, 1947), it was found that diisopropyl fluorophosphonate (DFP) or eserine sulfate could prevent the conduction block produced by prolonged exposure to isotonic solutions of acetylcholine chloride. Eserine 0.001 M was more effective than the same concentration of DFP, but DFP protection persisted after repeated washing in Ringer's solution and was correlated with persistent reduction in cholinesterase activity. Eserine prevented the irreversible effects of DFP exposure. The results illustrate typical anticholinesterasic actions of DFP and eserine in concentrations which were without demonstrable direct effect upon nerve, and further substantiate the lack of effect of unhydrolyzed acetylcholine upon frog nerve. In contrast to the anticholinesterasic actions of DFP and eserine, direct effects (increased threshold, decreased conduction velocity) at higher concentrations (0.025 M) were more pronounced but more quickly reversible with DFP than with eserine. The relation between conduction velocity v in m/sec and threshold i in arbitrary units was found to be: $v = L\ (2I - i)\ /IT$, where the constant I is threshold at moment of conduction failure, and L and T are the space and time constants of the fibers.

Effect of acetylcholine, caffeine, and alkaloids on activity of muscle adenosinetriphosphatase. Clara Torda and Harold G. Wolff. *New York Hospital and Departments of Medicine (Neurology) and Psychiatry, Cornell University Medical College, New York, N. Y.* The effect of acetylcholine and other chemical agents known to modify the shortening of muscle (alkaloids, caffeine) on the activity of muscle adenosinetriphosphatase was investigated. Actin-free myosin and actomyosin served as the source of the enzyme and adenosinetriphosphate (ATP) as the substrate (method of Szent-Györgyi and the modified method of Greenstein and Edsall as described by Singher and Meister). Acetylcholine, in low concentrations, increased the activity of the enzyme in splitting one phosphate from ATP. Caffeine, d-tubocurarine, and potassium, in concentrations corresponding to those inducing muscle contracture, decreased the activity of the enzyme. The other substances used (atropine, ephedrine, morphine, physostigmine, prostigmine, quinine, strychnine, and veratrine (in concentrations not interfering with the phosphorus determination)) did not modify the activity of muscle adenosinetriphosphatase. These results indicate that acetylcholine, by augmenting the activity of adenosinetriphosphatase, may contribute to a more adequate recovery of the muscle.

Experimental A-V block, method and discussion. William G. Turman (by invitation) and Jane Sands Robb. *Department of Pharmacology, Syracuse University, College of Medicine.* Desiring to record a-v bundle depolarization, sharp pointed shielded electrodes of silver chloride coated silver wire were fused into small glass tubes. These could be plunged into anterior or posterior septal regions or could be forced through the right ventricular wall and brought to rest near or upon the bundle

of His. In this additional position pressure usually produced temporary block. From direct surface or from indirect leads, potentials not reflected from the septal leads may appear and vice versa. In general, basal septal electrocardiograms have a more complex wave form than those from other heart areas. During supra ventricular rhythms septal P-R is shorter than P-R of indirect leads and septal activity precedes that of indirect leads. During block, with ventricular pacemakers, septal activity usually follows that of indirect leads. Records of one experiment with block are available where on lead 2 two ventricular pacemakers are shown but only one of these registers on the septal electrodes. These electrocardiograms from deep basal septal contacts probably show depolarization of the bundle of His; certainly show altered time relations when the pacemaker is ventricular; and suggest that not all ventricular premature beats spread to every area of ventricular muscle.

Blood, plasma and extracellular fluid volumes in experimental low colonic obstruction. A. H. TUTTLE (introduced by R. R. OVERMAN. *Department of Physiology, University of Tennessee, College of Medicine, Memphis, Tennessee.* Although previous investigators (Roberts and Crandall, Arch. Int. Med. **50:** 150, 1932) and most standard texts state that dehydration is an important factor in low colonic obstruction, modern methods of body fluid measurement have not been applied in this condition. Consequently, experiments have been performed using such methods to determine the nature and magnitude of body fluid changes in low colonic obstruction. Following complete experimental obstruction of the sigmoid colon in 9 dogs, simultaneous plasma volumes by the T-1824 dye method and "extracellular" fluid volumes by the thiocyanate dilution method were determined. Between the 16th and 24th post-operative days, the following average reductions in total volumes were found: plasma 6%, blood (calculated) 14%, extracellular fluid 16%, and erythrocyte mass (calculated) 25%. However, relative to changes in body weight, each of the above values with the exception of the erythrocyte mass was found to be increased. Since the extent to which the animals lost weight exceeded the extent of water loss, it is apparent that the primary weight reduction following low colonic obstruction is due to loss of tissue with low water content, presumably fat. More severe dehydration has been demonstrated in dogs during starvation, yet such reduction in body fluid volumes were not fatal, and the animals recovered when allowed food and water. We conclude that circulatory embarrassment and/or extracellular dehydration is not a serious sequel of low colonic obstruction

The effect of low thiamine intake on the reaction time of women. W. W. TUTTLE, MARJORIE WILSON (by invitation) and KATE DAUM (by invitation). *Departments of Physiology and Nutrition, State University of Iowa, Iowa City.* A group of 12 women ate a standard diet containing all nutritional requirements except thiamine. The standard diet did not contain more than 0.14 mg. thiamine per day. During a 6-week control period all the subjects ate the standard diet supplemented with adequate thiamine. During this period reaction times were measured so as to establish normal means for each subject. During a 6-week experimental period which followed, 6 subjects ate the supplemented diet, and 6 subjects ate the low thiamine diet. During this period measurements of reaction time were made. Reaction time to a light stimulus was measured with a Dunlap chronoscope. A comparison of the means shows that the reaction time of those eating low thiamine is significantly longer during the low thiamine period than during the period of adequate thiamine intake. The data also show that the reaction time of those on an adequate diet throughout the experiment did not become significantly longer. The data also support the conclusion that there was an increase in the dispersion of the accumulative frequency distribution of the reaction time of the subjects during the period that they were eating a diet low in thiamine.

The effect of nicotine on spinal synaptic conduction. A. VAN HARREVELD and GEORGE A. FEIGEN (by invitation). *California Institute of Technology, Pasadena, California.* In cats reflex action potentials were led off from the ventral roots of L7 or S1, while stimulating the homolateral dorsal root at the same segmental level. The injection of 5 mgm. nicotine per kg. bodyweight depressed or completely suppressed the monosynaptic spike, having a much smaller effect, which was usually temporary, on the multisynaptic activity. In agreement with this it was found that the kneejerk is suppressed, but that the flexion reflex is never abolished by such a doses of nicotine. Injecting more nicotine (up to 100 or even 200 mgm./kg.) caused the return of a monosynaptic spike which often became larger than before administration of the drug. The multisynaptic activity also was considerably increased. In preparations in which the peripheral curare-like effect of nicotine was prevented by establishing a separate circulation in the hind leg, it could be shown that the monosynaptic activity observed after large doses of nicotine is the equivalent of a myotatic reflex activity (kneejerk) like in the unpoisoned state. Nicotine, therefore does not seem to have the blocking affect on synaptic transmission in the spinal cord which it shows in sympathetic ganglia and in striated muscle. The depressing effect of the drug in small amounts on the kneejerk is probably due to a side action.

Effect of acclimatization on blood sugar re-

sponse to anoxia. EDWARD J. VAN LIERE, J. CLIFFORD STICKNEY and DAVID W. NORTHUP. *Dept. of Physiology, School of Medicine, West Virginia University, Morgantown.* Control dogs fasted for 24 hours were subjected to anoxic anoxia (254 mm. Hg) in a low-pressure chamber and blood sugar determinations made at the end of 15 minutes. Several such determinations were made at weekly intervals. After the control figure had been established, the animals were subjected to intermittent anoxia (303 mm. Hg) by keeping them in a low-pressure chamber four hours a day. Three of the four dogs showed acclimatization after a period of about 15 days since their blood sugar showed no significant elevation when exposed to a pressure of 226 mm. Hg for 15 minutes. One animal which was extremely sensitive to anoxia showed only partial acclimatization. To demonstrate that there actually is an acclimatization for every altitude and to show that the adrenal glands were not exhausted the acclimatized animals were subjected to a pressure of 208 mm. Hg for 15 minutes. Three of the four dogs showed a significant rise in blood sugar. The animal which showed only partial acclimatization died from anoxia. Deacclimatization was studied in the three remaining dogs. At weekly intervals they were subjected to a pressure of 254 mm. Hg and the blood sugar determined at the end of 15 minutes. One animal had lost most of his acclimatization after about 15 days; another started losing its acclimatization after about six weeks; another is still acclimatized although five months have elapsed since he was exposed to intermittent anoxia.

The role of the adrenal in the arterial pressure responses to severe hypoxemia. A. VAN LOO (by invitation), A. SURTSHIN (by invitation) and L. N. KATZ. *From the Cardiovascular Department, Research Institute, Michael Reese Hospital, Chicago, Illinois.* We have previously noted the effect of severe hypoxemia on the blood pressure of the dog. Breathing of pure nitrogen in open-chested dogs causes the development of an acute severe hypoxemia with a primary pressor phase, followed in a minute or so by a progressively falling blood pressure. When air breathing is resumed, the blood pressure rises rapidly to greater than control levels (posthypoxemic rise). We were interested in determining the factors responsible for these pressor responses, during the hypoxemic phase and in the posthypoxemic phase. In unilaterally adrenalectomized animals, diversion into a syringe of the venous blood draining from the remaining adrenal resulted in no significant change in the hypoxemic pressor phase, but diminished considerably the posthypoxemic response. Blood collected in the phase of falling blood pressure during nitrogen breathing had markedly more pressor activity than that collected during the earlier phases of hypoxemia. Reinjection of such collected blood immediately after the peak of the posthypoxemic response due to extra-adrenal factors caused the pressure to rise to a level similar to that attained in the control experiment. It is concluded that the adrenal gland plays little part in the production of the hypoxemic pressor response, but plays a major role in the production of the arterial pressor response after re-aeration. Pressor material liberated from the adrenal during severe hypoxemia does not exert its pressor effect until the tissues are re-oxygenated.

Post-fracture nitrogen loss with and without methionine supplements to good and poor protein diets. L. VAN MIDDLESWORTH and D. H. COPP (introduced by J. P. QUIGLEY). *Division of Physiology, University of California, Berkeley, and University of Tennessee, Memphis.* The effect of fracture upon daily urea + ammonia excretion was determined upon 6 groups of rats (6 animals per group) over periods of 20–90 days. The effects of the following synthetic dietary regimes were studied: (I) Complete diet containing 20% fibrin as the only protein. (II) Same as (I) but enriched with 2% dl-methionine. (III) Containing 10% casein as the only protein, otherwise complete. (IV) 9% casein plus 2% dl-methionine otherwise complete (same nitrogen content as II). Young adult female rats, which had stopped growing, were used with (I) and (II). Young adult males were used with (III) and (IV). The daily dietary intakes (9 gm. of diet/day) were limited such as to prevent growth. The animals weighed 175–200 gm. and their weights were maintained relatively constant over a 30-day control period. After single femur fractures the rats on (I) and (II) showed a cumulative loss of 200–400 mg. of nitrogen (above their basal excretion) during the first 8–10 days, after which their excretion returned to basal levels. The cumulative urea + NH_3 los of animals on diets (III) and (IV) was 200–280 mg. of nitrogen but this required 24–28 days. Sham operations on (III) and (IV) resulted in the cumulative loss of only 20–60 mg. of nitrogen and the increased excretion was not observed after 6–8 days. No consistant effect of methionine was demonstrated.

Vitamin A deficiency in pregnancy. G. VAN WAGENEN. *Dept. of Obstetrics and Gynecology, Yale School of Medicine, New Haven, Conn.* Normal pregnant monkeys and pregnant monkeys which had been less than a year on a diet deficient in Vitamin A were studied. The desquamation of keratinized epithelial cells as recorded in percentage of sediment in the vaginal lavage (method of Hartman) indicates ovarian activity. In young monkeys, before menarche, lavage yields few cells; while during the menstrual cycle vaginal contents may reach 50 to 100% density reflecting estrogen stimulation, then dropping at the

cycle end. During early pregnancy in the normal monkey desquamation remains at high levels as the many keratinized layers of superficial cells are shed just before, during and immediately after "implantation bleeding". In this first trimester the epithelium is reduced to a twentieth of its original thickness and appears as an inactive mucous membrane only a few cells thick. During the latter two-thirds of pregnancy, fluid from the vagina is clear and cell content extremely low. Paralleling this change in epithelial proliferation is the diminishing activity of the ovary.

In the A-deficient monkey keratinization of the vaginal epithelium persisted throughout pregnancy. This modification from the normal represents the imposed effect of vitamin A depletion common to many body epithelia and has been described in the pregnant rat by Mason ('35). The epithelial change here described in the monkey is probably an early digression from the normal at a low level of vitamin deficiency because it was compatible with the production of viable young showing no external developmental changes such as Warkany and Schraffenberger ('44) described in the rat.

Fractionation studies of enterogastrone activity in pyloric ligation rats. FRANK VISSCHER. *From the Research Laboratories of The Upjohn Company, Kalamazoo, Michigan.* We have submitted preparations of enterogastrone to electrophoretic separation at pH 7.5 and have tested the resultant fractions for anti-secretory activity in pyloric ligation rats, using light barbiturate anesthesia and a two-hour collection period. Seven fractions were each tested at three or more dosage levels to determine, by interpolation, the dose required for 50 per cent inhibition of volume of secretion. The fractions in order of increasing mobility gave the following approximate dosage requirements in mg. per kg. of rat: 6, 12, 23, 20, 18, 30, and 35. The most mobile fraction showed no inhibition at 20 mg. per kg. The original material had a dosage requirement of 10 to 15 mg. per kg.

We have also performed dialysis experiments involving three changes of external solution over a 72-hour period. About 50% of the material and nearly all of the remaining activity was recovered from the external solution. This material had an anti-secretory activity fully equal to that of the starting material, while the soluble fraction of non-dialysable material was nearly inactive.

The capacity changes in the pulmonary vascular bed with the respiratory cycle. MAURICE B. VISSCHER. *University of Minnesota.* Experiments were performed with isolated lungs from heparinized dogs to determine pulmonary vascular bed volume. Cannulae were fixed in the pulmonary artery and left atrium respectively, connected to calibrated tubes acting as recording reservoirs. The levels of fluid in the calibrated tubes were recorded by motion picture photography. The lung, cannulae and volume recorders were placed in a glass chamber, the tracheal tube was led out of the chamber and a second outlet was provided to alter the "intrathoracic" pressure. This system differs from those used previously by others, in that the head of pressure on the blood in the pulmonary vessels is always exposed to "intrathoracic" pressure. The earlier workers exposed the blood reservoirs to atmospheric pressure. Using the physiologically correct pressure head as described above entirely different results are obtained from those described by previous workers. It is found that the maximum blood capacity of the pulmonary vessels occurs at an "intrathoracic" pressure of -4.0 cm. H_2O. At more negative pressures the capacity of the lung vessels decreases progressively. For example, at -20.0 cm. H_2O in a 15 kg. dog with lungs weighing 108 gms. containing 50 cc. of expressible blood the decrease in lung vascular capacity was 2.3 cc. When the pulmonary vessel reservoirs are exposed to the same pressures as those acting on the lungs it is found that changes in "intrathoracic" pressure in the ranges of normal inspiration and passive expiration result in decreases in lung vessel capacity during inspiration, proportional to the degree of lung inflation, and increases during expiration.

Pupil dilatation in darkness as effected by intensity and duration of preexposure to white light. IRVING H. WAGMAN. *Department of Physiology, Jefferson Medical College, Philadelphia.* The course of pupillary dilatation in total darkness was determined for humans and rabbits following an exposure of two minutes duration to different brightnesses (1.6, 16, 52 footlamberts) of white light and following varying durations (15 seconds to 3 minutes) of exposures of a constant brightness. For the human, the visual field subtended an angle of 17° fixated centrally. The measurements were made by means of infrared photography. After the stimulus light is turned off, the pupil dilates rapidly for the first twenty seconds at which point it has reached more than half its maximum diameter. It dilates slowly from then on until it reaches its maximum size in about 6 to 10 minutes. The initial point of the pupillary dilatation curve and as a result, its subsequent course are determined by the intensity of preexposure. After exposure to a high intensity the pupil is smaller than after exposure to a lower intensity and therefore, the curve describing its course of dilatation is displaced to the right of the latter on the time axis. The curves, as far as can be determined are parallel. Varying the length of exposure of a preadapting light of constant intensity does not displace the curve. Such curves tend to follow similar courses. The course of pupillary dilatation seems to be determined, therefore, by the size of the pupil immediately after the light stimulus is removed.

The effects of percutaneous stimulation on the

circulation in normal and in paralyzed extremities. K. G. WAKIM, J. C. TERRIER (by invitation) and E. C. ELKINS (by invitation). *Rochester, Minnesota.* After control values for blood flow were established, the muscles in normal and in paralyzed extremities were stimulated percutaneously by the use of repeated condenser discharges at a frequency of about seventy stimuli per minute for a period of fifteen minutes. Determinations of blood flow were again taken at the end of the period of stimulation. The venous occlusion plethysmograph with a compensating spirometer recorder was used for the measurement of blood flow. Before and after stimulation of the extremities, skin temperatures were recorded galvanometrically. After electric stimulation of the spastic lower extremities of seven subjects, the blood flow in the stimulated extremities consistently showed an increase (in every one of the twenty-two experiments) which averaged 111 per cent over the control flow, with a range of plus 32 per cent to plus 340 per cent. The changes in skin temperatures were inconsistent; namely, in twenty-one experiments, the skin temperatures were reduced in three and increased in the rest. After stimulation, the spastic patients experienced a reduction in their spasticity which varied in magnitude and duration. Similarly, after stimulation the blood flow in normal lower extremities of eight healthy subjects consistently showed an increase which averaged 86 per cent, with a range of plus 10 per cent to plus 210 per cent. Normal subjects could not tolerate stimulation with the same intensity as that applied to paralyzed subjects. Of the eight subjects, six showed, after stimulation, a reduction in the skin temperature of the stimulated extremity, one showed no change, and one showed an increase.

Sixteen experiments were performed on the flaccid lower extremities of seven subjects who had lower motor neuron paralysis. After stimulation, four patients showed a reduction in blood flow, one showed no change, and the rest showed a slight increase; the average was plus 17 per cent and the range was minus 41 per cent to plus 48 per cent. Before and after electric stimulation the skin temperatures were recorded in 15 experiments, in nine they were decreased after stimulation, and in six, increased. The increase in blood flow occurring after electric stimulation concomitant with reduction in skin temperature in spastic and in normal extremities, and the insignificant increase in flow in atrophic flaccid extremities after stimulation, indicate that the increase in blood flow is contributed chiefly by activated muscles.

Interconversions of retinene and vitamin A *in vitro*. GEORGE WALD. *Biological Laboratories of Harvard University, Cambridge.* In intact retinas the bleaching of rhodopsin by light liberates the carotenoid retinene$_1$, which is subsequently converted to vitamin A$_1$. These processes can occur in the complete absence of oxygen, and in tissues poisoned with iodoacetate. Suspensions of isolated rod outer limbs, however, appear unable to convert retinene to vitamin A. In 1942 we prepared a cell-free brei from cattle retinas which accomplished this conversion. These experiments were interrupted by the War and have only now been resumed. Dark adapted retinas can be lyophilized, ground in a mortar, and extracted exhaustively in the dark with petrol ether. The residue, stirred up with neutral phosphate buffer, contains unaltered rhodopsin. On irradiation this bleaches, and the retinene formed is converted quantitatively to vitamin A. Recently the reverse conversion of vitamin A to retinene has been reported (Ball, Goodwin and Morton, Biochem. J., **40**, Proc. lix, 1946). Vitamin A alcohol, let stand in the cold for several days in contact with solid manganese dioxide, is replaced by a substance having a spectrum and yielding an antimony chloride test like those of retinene. We have confirmed these observations. We find that the adsorption of vitamin A by manganese dioxide plays an important part in the process. Retinene is less strongly adsorbed, and as fast as it forms it is displaced by new vitamin A. One can perform the conversion within a few minutes at room temperature, using a short column of manganese dioxide, pouring a petrol ether solution of vitamin A in at the top and drawing off a solution of retinene in the filtrate.

Further studies on the treatment and prophylaxis of experimental renal hypertension with renal extracts. G. E. WAKERLIN, JOHN MARSHALL, HIROAKI MINATOYA, RUFUS WALKER and A. KAPLAN. *Department of Physiology, University of Illinois, College of Medicine, Chicago.* In further work to determine the mechanism of the antihypertensive effect of hog renal extracts and to obtain a consistently potent, purified extract, we studied 2 new crude hog renal extracts in the treatment of renal hypertensive dogs and 5 fractions of one of the crude extracts in the prophylaxis of experimental renal hypertension in dogs. One of the crude extracts was prepared as previously described, except that autolysis was minimized by prompt acetone treatment at −20°C. Daily intramuscular injections of this extract for 10 months produced only a fair antihypertensive effect and high serum antirenin titres (10 and 16 units per cc.) in 2 renal hypertensive dogs. Treatment with the second extract prepared from Viobin processed kidney powder for 5 months gave no antihypertensive effect and no antirenin in 2 hypertensive dogs and a fair effect and modest antirenin titre (2 units) in one animal. Five ammonium sulfate fractions were studied prophylactically. Only one dog was protected against chronic experimental renal hypertension, 8 animals developed chronic hypertension similar to 11 of the 14 controls, and 2 dogs developed renal insufficiency without hyper-

tension and without arteriolonecrosis in contrast to 3 of the controls which died in typical malignant hypertension. The antirenin titres of the treated animals varied from 0.4 to 22 units, with 5 unit titres for the protected dog and the two animals showing fatal renal insufficiency without malignant hypertension.

The data do not permit conclusions relative to the renin-antirenin mechanism as operative in the modest therapeutic and prophylactic effects observed. The results suggest that preliminary renal autolysis may be of significance in determining antihypertensive extract potency.

The relationship between anticoagulants and lipemia. J. M. WALDRON (by invitation), and M. H. F. FRIEDMAN. *Department of Physiology, Jefferson Medical College, Philadelphia.* Hahn (1943) and Weld (1944) reported that a small dose of heparin intravenously cleared alimentary lipemia within five minutes and that this phenomenon did not occur in vitro. We found that heparin not only clears alimentary lipemia but also lipemia of carbon tetrachloride poisoning. Furthermore, other anticoagulants (a synthetic heparin-like sulfonated polysaccharide and pontamine fast pink B.L.) have the same effect. In addition to clearing alimentary lipemia heparin has a reverse effect. If corn oil is administered in an amount which, in itself, does not produce lipemia, the blood will become lipemic within five minutes after the intravenous injection of heparin or the other anticoagulants. This lipemia-potentiating effect of anticoagulants does not occur in vitro or without prior administration of fat. Thus an attempt has been made to utilize this property of heparin as a test for intestinal absorption of fat. As used by previous authors and by us the term "lipemia" refers only to the turbidity of the serum or plasma without reference to the quantity or physical state of the fat. The turbidity returns to the heparin-cleared serum if left in a test tube at room temperature for eighteen hours, whereas the lipemia induced by heparin does not clear under the same conditions. The occurrence of both the lipemia-clearing and lipemia-potentiating effects are correlated with blood coagulation time. The anticoagulant effect of heparin is modified by the ingestion of fat.

Action potentials induced by indirect stimulation of rat muscle after adrenalectomy, KCl treatment and tetanus. SHEPPARD M. WALKER. *Dept. of Physiology, Washington University School of Medicine, St. Louis, Mo.* The records were obtained under ether anaesthesia from immature rats. Action potentials were recorded from the gastrocnemius muscle with a cathode ray oscillograph. Mechanical records of muscle contraction were made simultaneously using an isometric lever and optical recording. The cut sciatic nerve was stimulated with brief shocks 3 to 4 times threshold strength. The records from adrenalectomized animals were obtained after the appearance of signs of severe adrenal cortex insufficiency. Normal rats were injected intraperitoneally with 40 to 80 mgm. of KCl per 100 grams of body weight. Tetanus was produced by repetitive shocks at the rate of 200 per sec. for 1 to 2 seconds. Control records were made from rested muscle of normal rats.

The mechanical responses to single stimuli were larger in muscle of adrenalectomized, KCl-treated or tetanized rats than in muscle of control animals. The action potentials which accompanied the large mechanical responses showed no electrical activity which could be separated from the initial action potentials either when both lead electrodes were placed in the belly or when belly to tendon leads were employed. The duration of action potentials was longer in muscle after adrenalectomy, KCl treatment or tetanization than in control muscle. The height of action potentials was reduced by KCl treatment and by tetanization. The results indicate that the increase of mechanical response of adrenalectomized, KCl-treated and tetanized muscle to single stimuli is due to increased contractile strength of the muscle rather than to repetitive response of a part of the muscle fibers.

Effect of secretin and pancreozymin on amylase and alkaline phosphatase of dog's pancreas. C. C. WANG (by invitation) and M. I. GROSSMAN. *Dept. of Clinical Science, University of Illinois College of Medicine, Chicago.* Highly purified secretin dissolved in saline was constantly injected by means of a perfusion pump into the femoral vein of anesthetized dogs with a cannula in the major pancreatic duct. Increase of flow was brought about by increasing the rate of injection of secretin. At least three or four samples of pancreatic juice were collected at each secretory rate level. Pancreozymin was injected in a single dose of 20 mg. during the response to a constant dose of secretin. All the samples were assayed for amylase and alkaline phosphatase content.

It was clearly shown that secretin had no stimulant effect on enzyme production, neither amylase nor alkaline phosphatase. As the volume of pancreatic juice was increased after increasing the rate of secretin injection, the concentrations of both of these enzymes were decreased, and the amounts of enzyme discharged per minute remained essentially unchanged in case of amylase and decreased in case of alkaline phosphatase. Pancreozymin, however, increased the concentration of amylase in the juice three or four-fold, but the alkaline phosphatase remained unchanged. This suggests a very significant point; namely, that the source of enzyme production is probably not the same for phosphatase as for the other enzymes in the pancreas. We propose that the ductule cells are responsible for the production of the phosphatase and the acinar cells for the other three enzymes, namely amylase, trypsinogen and lipase. Histo-

chemical demonstration of alkaline phosphatase in pancreatic tissue of the dog supports this proposal.

Serum accelerator globulin: quantitative determination, purification and properties. ARNOLD G. WARE (by invitation) and WALTER H. SEEGERS. *Department of Physiology, Wayne University, College of Medicine.* Following the discovery of a factor in plasma which accelerates the interaction of prothrombin, thromboplastin, and calcium ions, we found that a similar factor is present in serum but it is far more potent in its accelerator action (Ware, Murphy and Seegers, Science, in press). We refer to the plasma factor by the term, plasma Ac-globulin, and to the serum factor by the term, serum Ac-globulin. Thrombin produces serum Ac-globulin from plasma Ac-globulin, apparently by catalytic action. Plasma Ac-globulin Thrombin Serum Ac-globulin. Extremely small amounts of thrombin will produce the change. Larger amounts of thrombin have a similar action but in addition actually destroy the accelerator activity. The chemical properties of serum Ac-globulin are so similar to the properties of plasma Ac-globulin that the same method is used for purification as that described for the plasma accelerator factor (Ware and Seegers, J. Biol. Chem., in press). Serum Ac-globulin has been obtained in concentrated form both from serum and from defibrinated oxalated plasma. It has also been obtained in high yields from defibrinated oxalated plasma after storage for 22 days at 5°C. This fact and other data indicate that serum Ac-globulin is relatively stable in serum. Quantitative measurements of its activity depend upon its ability to accelerate the interaction of prothrombin, thromboplastin, and calcium ions. This is done by an adaptation of the 2-stage prothrombin analysis.

Influence of tea from leaves of Ceanothus americanus on blood pressure of hypertensive rats. HELENE WASTL. *Dept. of Anatomy, Hahnemann Medical College, Philadelphia.* Leaves of Ceanothus americanus (Rhamacea family) have been used widely as a satisfactory tea-substitute during the American Revolutionary War. The taste of this popularily called "New Jersey tea" is slightly more astringent. In 1926 Ceanothin, alcoholic extract of the root bark from the shrub has been shown to have marked blood pressure and clotting time reducing properties. The tea was tested on hypertensive rats, which were fed it once daily by hand.

Mm. Hg systolic blood pressure

	Normal	Hypertension	New Jersey tea feeding	Cessation of tea feeding	Reduction of hypertension
Average.....	126	168	132	168	−36
Range.....	108-140	128-230	103-163	128-230	−3 to −80
Average.....	126	182	147	185	−35
Range.....	110-140	154-230	119-171	160-230	−16 to −62

During tea-feeding-periods (first group, 18 rats, for 12 days; second group, 18 rats, for 24 days) reduction of hypertensive blood pressure increased in steps up to 6–8 days, when the final level characteristic for each individual rat is reached. Lengthening of tea-feeding periods does not achieve better results. The values refer to the whole pre-feeding, feeding and after-feeding periods averages; blood pressures were measured every 4 days. G. W. Boericke, Dept. of Therapeutics, Hahnemann Medical College, observed recently in a number of hospitalized cases of essential hypertension also encouraging reductions of blood pressure levels under New Jersey tea medication. This study, on animals as well as on patients, will be continued.

Regeneration of insulin fibrils with several reagents and the nature of the inter-insulin bond. DAVID F. WAUGH, M. JANETTE SMITH (by invitation), and DARTHEA F. FEARING (by invitation). *Dept. of Biol., Mass. Inst. of Tech., Cambridge.* Insulin fibrils (Waugh, J. Am. Chem. Soc. **66**, 663, 1944) may be regenerated yielding products resembling insulin in crystallization, biological activity, and fibril formation. Some compounds producing regeneration are: (1) Acids: 40% sulphuric acid at minus 10°C., and 38% hydrochloric acid at plus 5°C. give crystalline yields of 50%. (2) Alkalis: 0°C. to 15°C., 0.03N, and times of 5 to 10 minutes give 90% yields. (3) Liquid ammonia: minus 70°C. yields 90–95%. (4) Phenol: regenerates over wide ranges of temperature and concentration giving maximum yields above 90%. Several compounds such as nitrobenzene, chlorobenzene, and pyridine do not regenerate fibrils themselves and inhibit regeneration with phenol. After treatment with such compounds fibrils may be recovered and subsequently regenerated with phenol, thus showing that they do not regenerate and destroy the regeneration product. Salts, i.e. 0.1N sodium chloride and sodium acetate, inhibit regeneration with those compounds tested (2 and 4) indicating that repulsive forces between similarily charged particles are important. This agrees with the inability of phenol to regenerate over a 3 pH unit range near the isoelectric point. At 60°C. and fibril concentration of 0.4%, 10–12% phenol regenerates rapidly. In 5% phenol fibrils will elongate in the presence of native insulin. An attempt will be made to show that these observations support a conclusion drawn previously (loc. cit.) that the inter-insulin bond is due to the coalescence of hydrophobic residues.

Leucocytosis of guinea pigs deficient in the anti-stiffness factor. VIRGINIA WEIMAR (by invitation) and ROSALIND WULZEN. *Dept. of Zoology, Oregon State College.* Guinea pigs fed a skim milk diet, supplemented by the known vitamins and necessary minerals but deficient in the anti-stiffness factor, developed a characteristic wrist stiffness. Autopsies showed the muscles to be finely streaked with calcium deposits running parallel to the

muscle fibers. Large deposits of tricalcium phosphate were frequently found along the body wall, around the joints, and in numerous organs (Wulzen *et al.*, Am. J. Physiol. 133, 500, 1941). Leucocyte enumerations were made on 26 guinea pigs which had been fed for 15 to 98 weeks on the diet deficient in the anti-stiffness factor. These animals had white counts of 11,000 to 30,000 (mean 17,668). Fifty-five young and adult male and female guinea pigs on a grain and kale diet had leucocyte counts of 4,000 to 11,900 (mean, 7,459). The "Student's" t-test was used to demonstrate the difference in the means of the two groups. The t was -14.33; degrees of freedom 79. These results indicate that deficiency of the anti-stiffness factor in guinea pigs produces a significant leucocytosis.

The effect of serum hypersensitivity reactions upon the electrocardiogram. GRACE E. WERTENBERGER and ROBERTA HAFKESBRING. *Department of Physiology, Woman's Medical College of Pennsylvania, Philadelphia, Pennsylvania.* During an investigation of the effects of the sulfonamides upon the myocardium, electrocardiographic changes indicating alterations in the physiologic state of the muscle as shown by prolonged conduction time, ST segment deviation and change in the direction of the T wave were found in 15% of the animals. No electrocardiographic changes occurred with the initial course of the drug, even in animals whose controls showed prolonged conduction time, heart block and ST segment shifts indicating established cardiac pathology. The cardiac changes appeared with subsequent courses of the drug and, although transient in some and persistent in others, were always associated with severe drug reactions.

Since the side reactions to the sulfonamides suggest hypersensitization with the drug acting as the sensitizing antigen, the present study to compare the functional effects of serum sickness and sulfonamide hypersensitivity upon the myocardium was begun. In the first series of serum sensitized rabbits, the electrocardiogram showed only asphyxial changes in the animals dying of anaphylactic shock and no significant changes have as yet appeared in the three animals that survived the shocking dose. The second series is now being followed. In the series of serum sensitized dogs now being studied, changes have appeared in the electrocardiogram which cannot be attributed to asphyxia.

The renal excretion of strong electrolytes. LAURENCE G. WESSON, W. PARKER ANSLOW, JR. AND HOMER W. SMITH. *New York University College of Medicine.* Indirect evidence (Smith, Oliver *et al.*) indicates that roughly 85 per cent of the sodium and water in the glomerular filtrate are reabsorbed by the proximal tubule and thin limb in the mammalian kidney, and that tubular urine is isosmotic with plasma half way down the proximal tubule (Oliver *et al.*). Further quantities of sodium and water are reabsorbed in the distal tubule by independent processes. Observations on dogs during osmotic diuresis induced by mannitol reveal that up to 65 per cent of the water of the glomerular filtrate may be excreted with a simultaneous excretion of only 27 per cent of the filtered sodium. The sum of the osmotic constituents in the urine remains practically identical with the plasma. It is concluded first, that the proximal reabsorption of sodium and water are at least partially independent processes and second, that proximal water reabsorption is a process of passive diffusion initiated by the active reabsorption of sodium, with its attendant anions. Passive osmotic equilibration may be completed in the thin limb so that the urine delivered to the distal tubule is normally isosmotic with plasma. If the limiting values in either sodium or water reabsorption in the distal tubule can be determined experimentally, the partition of both constituents between proximal and distal reabsorption can be calculated.

Tentative evidence is adduced that under the action of ADH distal reabsorption of water reaches a maximal rate ($T^d_{m_{H_2O}}$) and that under conditions conducive to stability the distal reabsorption of sodium is also limited by a maximal rate ($T^d_{m_{Na}}$). Under such conditions sodium excretion is conditioned by the filtration rate as well as the plasma level of sodium.

Effect of low sodium and Kempner diets on renal hemodynamics and electrolyte excretion in hypertensives. RAYMOND E. WESTON, LEON HELLMAN (by invitation), DORIS J. W. ESCHER (by invitation), and LOUIS LEITER (by invitation). *Medical Division, Montefiore Hospital, New York 67, N. Y.* The Kempner rice diet, which has been recommended for the treatment of hypertension, provides an opportunity to follow changes in renal hemodynamics and electrolyte excretion during severely restricted sodium and protein intake. Therefore, in selected hypertensive patients without demonstrable specific renal pathology, glomerular filtration rates, renal plasma flows, maximal tubular PAH excretory capacities, and sodium, chloride, and urea excretions were studied on normal diets, after several weeks on low sodium (0.8) but normal protein (65 grams) diets, and after four to sixteen weeks on the Kempner (0.2 grams sodium and 30 grams protein) regime.

Preliminary observations indicate that in hypertensives, the low salt diet produces a decrease in filtration fraction, since there is a significant decrease in GFR with a lesser change in RPF, and no change in $T_{m_{PAH}}$. In some patients, the more severe sodium restriction and/or the reduced pro-

tein of the rice diet result in a further decrease in GFR, a greater decrease in RPF, and some diminution in T_{mPAH} As the diets were varied, the changes observed in electrolyte excretion were as expected, with certain exceptions which apparently are characteristic of the hypertensive kidney. In this small series, none of the patients exhibited a significant drop in blood pressure while on either the low salt or the rice diet.

Further experimental analysis of these results is in progress, with particular emphasis on the influence of plasma volume, extracellular fluid volume, electrolyte and protein metabolism, and endocrine factors.

Effects of exercise on renal circulation. H. L. WHITE and DORIS ROLF (by invitation). *Dept. of Physiology, Washington University School of Medicine, St. Louis, Mo.* The effects of exercise on para-aminohippurate (PAH) and inulin clearances (subcutaneous administration), on venous plasma glucose, mean blood pressure (diastolic plus 40% pulse pressure), renal vascular resistance, and urine protein were determined in six normal human males. Blood pressures were not obtained during exercise. Control periods were during recumbency. Light exercise (walking alternating with slow running for 15 minutes, or slow running for 12 minutes) produced a slight fall or no change in PAH clearance, with slight further falls in first two post-exercise periods; there was no significant change in filtration fraction.

Heavy exercise, to exhaustion, panting and profuse sweating, by running at maximum capacity for 12 minutes, gave great falls (to 20% of normal) in PAH and inulin clearances during exercise; the values had not returned to normal within 60 minutes. Filtration fractions showed no consistent changes during and after exercise. Renal vascular resistance increased at least 5 fold (taking mean arterial pressure during exercise as normal) during exercise and was still somewhat above normal an hour later. All urines were protein-free before and *during* heavy exercise, but urines formed during first and second post-exercise periods contained protein, which had disappeared by the third 20 minute post-exercise collection. This supports but does not prove the view that during heavy exercise many glomeruli may have no blood flow. Rise in plasma glucose, probably indicative of adrenalin output, was slight or absent on light, but up to 100 mgm. % on heavy exercise.

Circulatory responses to exposure to barometric pressure of 30 mm. Hg. WILLIAM V. WHITEHORN. *Dept. of Physiology, Ohio State Univ.* Nembutal anesthetized dogs explosively decompressed from 560±5 to 30±5 mm. Hg show drops in arterial pressures initially similar to those seen following explosive decompression to less extreme pressures. Recovery to normal levels during exposure to the reduced pressure does not occur, however—mean pressures falling to an average level of 70.7 mm. Hg 30 sec. after decompression and remaining essentially unchanged for periods up to one minute. Arterial pressure curves indicate marked diminution in pulse pressures or actual disappearance of arterial pulses within 30 seconds after decompression although e.c.g. records show continuing electrical cardiac activity. Observations on exposed arteries and veins under these conditions reveal massive venous but absent arterial intravascular bubble formation. The described phenomena do not occur with decompressions to final pressures greater than about 50 mm. Hg. It is postulated that the circulatory changes described are the result of blocking of veins and pulmonary vessels by gas formation with resulting reduction or absence of left ventricular output. The maintenance of mean arterial pressure at a constant low level under these conditions is not entirely clear but may be the result of increased tissue pressures due to interstitial gas formation at final pressures less than 47 mm. Hg.

Rhesus hyperkinesia by subthalamic lesion. JOHN R. WHITTIER introduced by FRED A. METTLER. *Department of Neurology, Columbia University, College of Physicians and Surgeons.* (Motion Picture). A variety of abnormal involuntary movements have been produced in rhesus monkeys by small electrolytic lesions placed in the region of the subthalamic nucleus with stereotaxic technique. Choreoid, athetoid, and choreo-athetoid patterns appeared upon recovery from anesthetic and persisted in most cases until sacrifice, as long as two months after operation. They varied among the animals from the appearance of irregularly alternating movements at a single joint to such forceful sequences in axial and appendicular muscles that hips and limbs were flung about in a violent manner and the act of feeding became a hectic conflict between voluntary and involuntary activity. Unequivocal hyperkinesia appeared in 20 monkeys of a series of 80, and in every instance the subthalamic nucleus was involved by the lesion. In 37 animals for which histological preparations are available to date the subthalamic nucleus was damaged in 11 who showed no hyperkinesia. Tentative analysis suggests that in these cases the involvement of the nucleus in the lesion was small, or extension by the lesion into surrounding structures was considerable.

Lantern slides illustrate the lesions and a motion picture demonstrates varieties of hyperkinesia obtained.

Preparation of anti-ulcer substance from bovine urine. ARNE N. WICK and FRANCES PAULS (introduced by EATON M. MACKAY). *Scripps Metabolic*

Clinic, La Jolla, Calif. The preparation of an anti-ulcer factor from human urine by charcoal adsorption has previously been reported (Wick, Irish, Pauls, and MacKay, Proc. Soc. Exper. Biol. and Med., **64**: 40, 1947). Examination of cow urine (obtained from milking herds 75 per cent of which are customarily bred) for this factor disclosed an excellent source for a non-toxic anti-ulcer preparation. This material can be obtained by treating the urine with carbon followed by elution of the active principle with aqueous acetone. Crude dialyzed samples obtained by this method completely prevented gastric ulceration in the rat assay (Pauls, Wick, and MacKay, Gastroenterology, **8**: 774, 1947) when administered intravenously at a dose level of 100 mg. per kilogram. Chorionic gonadotropic assays in the immature rat gave a negative response at 1 I.U. per mg. of test sample. The benzoic acid procedure (Gray, Wieczorowski, Wells, and Harris, Endocrinology, **30**: 129, 1942) when applied to bovine urine, in contrast to human urine, is not suitable for the isolation of the ulcer factor (urogastrone) because of the apparent presence of a solubilizing agent.

The effect of body temperature on the respiratory transformation of intravenous carbohydrates in amytal-anesthetized dogs. M. WIERZUCHOWSKI (introduced by WILLIAM H. CHAMBERS). *Physiological Institute of the Medical Faculty, University of Lodz, Poland.* Moderately deep amytal anesthesia was maintained in dogs during continuous infusion of sugars at constant rate of 40 g./sq.m./hr., with rectal temperature varying from 33.7–41.8°C. Total utilization of carbohydrate, as well as ventilation and oxidation, increased with the rise of body temperature; the ratio of ventilation/O_2 consumption and heat production above basal grew constantly. Specific dynamic action of glucose, fructose, maltose and galactose, referred to their total utilization, increased with body temperature according to a straight line equation. The curves run parallel and fairly closely to each other (e.g., for glucose, 4% at 35°C., 13% at 40°C.) similar at a normal range to those of unanesthetized dogs. Intravenous glucose at rates of 30–214 g./sq.in./hr. was supplied to amytalized dogs for various ranges of body temperature. When plotted against blood glucose concentration, similar curves were obtained for total utilization, ventilation, O_2 consumption, CO_2 elimination and heat production. The curves rose at an exponential rate to a peak value which remained constant up to 4000 mg.% of blood glucose. The peak value was about 1000 mg.% of blood glucose when the body temperature fell within normal limits. In hypopyrexia the peak was lower than 1000 mg.% and in hyperpyrexia it was higher. At the peak value glucose oxidation was ½ of total assimilation, as in the unanesthetized animal (e.g., above 39°C., with a blood sugar of 1200 mg.%, 20 g./sq.m./hr. were oxidized out of 100 g. assimilated). Amytal anesthesia does not seem to effect carbohydrate transformation except when changes occur in body temperature. Body temperature appears to be a factor of great importance in the consideration of the specific dynamic action of intravenous carbohydrates.

Glucose metabolism in marine invertebrates. CHARLES G. WILBER. *The Biological Laboratory, Fordham University, New York.* There is practically no information about metabolism in marine annelids to be found in the literature. Some studies have been made on the effect of starvation on the lipids in *Phascolosoma* (J. Cell. and Comp. Physiol., **29**: 179, 1947). A series of experiments was, consequently, made to ascertain the effect of increased temperature on glucose metabolism in the annelid, *Phascolosoma gouldii*. Body fluid was removed from 12 specimens, which had been kept at 20°C. for several days, and analyzed for glucose. Using the Folin method an average of 17.3 mg. glucose/100 cc. fluid was found. Using the Somogyi filtrate 4.6 mg./100 cc. glucose was found. A second dozen phascolosomas were put into a salt water bath at 30°C. for 30 minutes after which the body fluid was analyzed. With the F. method the glucose value was 30.2 mg./100 cc.; with the S. filtrate, 26.1 mg./100 cc. The differences are statistically significant. Apparently, the increased temperature stimulated the breakdown of glycogen, stored in the muscles or elsewhere, to form glucose. The ultimate fate of the large amount of glucose is now being studied with the view to throwing some light on the carbohydrate metabolism of marine invertebrate animals.

Relationship in the dog between inferior vena caval pressure and total plasma protein. J. LARUE WILEY, MICHAEL NEWTON and J. B. TRACY, JR., (introduced by H. C. BAZETT). *Department of Physiology, Medical School, University of Pennsylvania, Philadelphia, Pa.* Utilizing changes in the ratio Total Hb/Total Protein = Hb concentration/Plasma protein concentration (1 − hematocrit) as indicating fluctuations in total plasma protein, (Spealman, Newton and Post, Am. J. Physiol. **150**: 628, Oct. 1947) it was found in human subjects that total circulating plasma protein increased over periods of several hours when posture was changed from recumbent to erect. The effect on the ratio Hb/Protein, of artificially increasing venous pressures below the heart was studied in acute experiments on dogs. All animals were first splenectomized to eliminate at least one variable reservoir of erythrocytes. The inferior vena cava was constricted either between the diaphragm and heart, or between the renal veins and liver, resulting in elevation of distal caval pressure by 100–200 per cent. Control animals underwent the same operative procedures but had no constriction ap-

plied to the vena cava. Results in 7 experimental animals compared to 8 controls were inconclusive. There was a slight downward trend in the ratio Hb/protein when the thoracic inferior cava was constricted. However total circulating hemoglobin may have changed somewhat under conditions of these experiments, and the magnitude of changes in ratio do not justify the assumption of changes in total protein. Chronic preparations now being studied may yield more positive results.

The asystolic arterial pressure gradient as a measure of local peripheral resistance. ARNOLD H. WILLIAMS (by invitation) and HENRY A. SCHROEDER. *Department of Internal Medicine and the Oscar Johnson Institute, Washington University School of Medicine, and Barnes Hospital, St. Louis, Missouri.* The hemodynamics of any artery may be simplified and likened to an expansile reservoir in which the pressure is principally regulated by the relative rates of cardiac input and arteriolar drainage. When the input is suddenly checked, as in diastole, the intraarterial pressure is maintained in part by the elastic recoil of the arterial wall but to a much greater extent by the rate of outflow into the capillary bed. The outflow rate depends on the caliber of the arteriolar "stopcock" which is the principal site of peripheral resistance. It follows that the gradient of blood pressure fall, distal to the point of a sudden occlusion of an artery, is a measure of the outflow or peripheral resistance, and that the arterial pressure approximates the local venous pressure with prolonged occlusion. This principle has been employed in dogs and man as a measure of peripheral resistance in various vascular areas. After arterial occulsion at the peak of systole the subsequent intraarterial pressure gradient has been determined with the Hamilton manometer in various end arteries such as those supplying the limbs, the mesentery, and the kidney. It is probable that this technique can not be utilized as a quantitative measure of peripheral resistance. However, it is extremely useful in following the duration and site of local vasoconstrictor action after the injection of pressor and depressor agents. The changes that occur are exaggerated by logarithmic plotting of the pressure gradients. Hemodynamic analyses of the factors involved and mathematical analyses of the gradients obtained are in progress.

Sodium, potassium and chloride in leucocytes. D. L. WILSON (by invitation) and J. F. MANERY. *Department of Biochemistry, University of Toronto, Toronto, Canada.* Rabbit leucocytes were equilibrated in Ringer-Dale solutions which contained concentrations of chloride varying from 25 to 160 m. eq. per litre; by substituting appropriate amounts of sodium sulphate for sodium chloride the solutions were made isosmotic. Suitable tests showed the leucocytes to be viable in all solutions used. Packed cells, obtained by centrifugation, were analysed for sodium, potassium, chloride and water. The data indicated that intracellular chloride concentrations varied directly with those outside, always being lower at external concentrations above 55 m. eq. per litre. Evidence of an anion exchange was obtained. Although the analytical methods used for the determination of sodium and potassium gave results reproducible to $\pm$ 1%, the intracellular concentrations varied over a range of $\pm$ 20%. Under any set of conditions, however, the sum of the potassium and sodium concentrations remained constant suggesting that a reciprocal relation existed between the two cations. After aerating gently for 45 minutes in a Ringer-Dale solution containing 115 m.eq. of chloride per litre (37°C.), the leucocytes contained 80% water, 80 to 90 m.eq. of chloride, 65 to 100 m.eq. of sodium and 75 to 110 m.eq. of potassium per l. of cell water. These figures imply that factors other than the external concentration of sodium and potassium influence the concentration of these cations within the cells.

Comparative properties of six phenethylamines. CLAUDE V. WINDER, MONA M. ANDERSON (by invitation) and HERVEY C. PARKE (by invitation). *Research Laboratories, Parke, Davis & Co., Detroit, Michigan.* The hydrochlorides of phenethylamine, its α- and β-methylated derivatives, and the three corresponding N-methyl compounds (racemic forms in all cases of asymmetry) were studied along with *l*-ephedrine in phenobarbitalized, atropinized dogs. Average molecular pressor potencies (46–500 μgm./kgm. dose level) ranged between a few and several thousandths of epinephrine's. The β-methylated compounds were significantly less potent. Intravenous nasal-paranasal decongestant action was recorded volumetically (special miniature spirometer). Decongestant efficiency was defined as the ratio of decongestant potency to pressor potency as determined against respective epinephrine scales from a common set of injections —an efficiency index with epinephrine set at unity. The parent amine and its N-methyl derivative were least efficient (significantly less than epinephrine), the β-methylated intermediate, and the α-methylated most. In general, the primary amines were more efficient than the secondary. Inferences concerning nasal *vasoconstrictor* efficiency bear reservations. Durability of action was measured in terms of time from beginning of effect until recession to half-value. Its ratio to the same measure of the equipressor epinephrine response was termed the 'duration index' and was satisfactorily independent of dose level. Tachyphylaxis was studied systematically by repeated injections at decreasing time intervals of *ca.* equipressor doses (174–571 μgm./kgm.). The non-chain-methylated amines were least, the J-methylated intermediate, and the

α-methylated most durable and tachyphylactic. *l*-Ephedrine was somewhat less durable and less precipitously tachyphylactic than the non-hydroxylated isopropylamines.

On the nature of tachyphylaxis and related phenomena. CLAUDE V. WINDER and MONA M. ANDERSON (by invitation). *Research Laboratories, Parke, Davis & Co., Detroit, Michigan.* Tachyphylaxis of *dl*-amphetamine, *dl*-desoxyephedrine and *l*-ephedrine hydrochlorides, at *ca.* equipressor dose levels, was studied in arterial pressure and nasal (de) congestion (calibrated volume recorder) of phenobarbitalized, atropinized, vagotomized dogs. In each experiment one drug was repeatedly injected at intervals decreasing from 1 hr. to 10 min. Tachyphylaxis in pressor and decongestion effects were strikingly parallel. While blood pressure more or less completely returned to 'normal' between injections, successive decongestions of the neurally isolated mucosa were relatively well-sustained and correspondingly additive. Plots of *cumulative* peak decongestion, as percent. of 'total possible decongestion' on a probability scale, against logarithm of *cumulative* dose, yielded amazingly satisfactory linear fits in all cases. Hypotheses met by such plots are: (a) the dynamic proportion of a population of 'receptor points' occupied by the agent is of the nature of a statistical (probability) cumulation according to the agent's concentration; (b) the measured end effect is analogous to the pyrimiding of successive orders of statistical samples between initial physico-chemical event and final functional change; (c) the statistical distribution is log-normal with respect to the agent's concentration; (d) a series of tachyphylactic responses reflect successive (more or less overlapping) stages of cumulative probability of receptor occupation; (e) the cumulating factor (in equal arithmetic, diminishing logarithmic steps) may be more (blood pressure) or less (mucosa) compensated between administration. Computations predict that tachyphylaxis, potentiation, and certain passive 'interferences' may be in important degrees kaleidoscopic manifestations of (a) compactness of probability range of agent-receptor engagement, (b) dose, (c) disposal, and (d) number of previous injections.

Gastrointestinal and hematological responses in dogs to large doses of histamine and an antihistaminic drug. CHARLES A. WINTER and CHARLES W. MUSHETT (by invitation). *Merck Institute for Therapeutic Research, Rahway, N. J.* Previous workers have produced experimental peptic ulcers in dogs by prolonged histamine administration. However, when an animal is protected against the acute lethal effect of histamine by an antihistamine agent, relatively large doses of histamine may be given, thus reducing to a few days the time required to produce ulcers. Antihistaminic drug (Neo-antergan) was administered 10 mg./kg. in aqueous solution subcutaneously one-half hour before histamine, followed by another dose intramuscularly in oil at the time of histamine administration. Each of 3 dogs received a single injection of histamine in oil-beeswax, 20 to 40 mg./kg. intramuscularly. Sacrifice 48 hours later revealed denudation or discrete ulceration in the duodenum in all 3. Typical ulceration was seen in 8 dogs receiving 2 to 5 such doses. In this group, sacrificed 3 to 7 days after the first dose, all animals had duodenal ulcers, and 7 had gastric ulcers.

Hemoconcentration occurred regularly within 2 to 4 hours following an initial injection of 40-50 mg./kg. of histamine, and was still apparent, though decreased, after 24 hours. Initial leukopenia occurred within a few hours, but was sometimes masked by the coexisting hemoconcentration. However, within 24 hours a leukocytosis of variable degree usually developed, which often became more marked after additional histamine injections. Most animals thus showed a 2 to 4 fold increase in total leucocyte count. An increased erythrocyte sedimentation rate occurred frequently within 24 hours. These data indicate that the antihistaminic drug prevented neither the gastrointestinal lesions nor the hematological effects produced by histamine.

The transport and excretion of uric acid in man. IV. The renal mechanism for urate excretion. W. Q. WOLFSON (by invitation) and R. LEVINE. (*From the Department of Biochemistry and the Department of Metabolic and Endocrine Research, Medical Research Institute, Michael Reese Hospital, Chicago.*) A major obstacle in the study of human urate excretion has been the difficulty of provoking hyperuricemia in normal subjects without the toxicity accompanying i.v. administration of urate. This may be overcome by administering large fractional doses of nucleate orally on the day preceeding the clearance. Administration terminates at midnight and clearances are determined on a falling curve. Urate levels up to 12.5 mg.% were attained. At an average plasma urate of 6.26 mg.%, the clearance represented 8.3% of the glomerular filtration rate. At 10.06 mg.%, the urate clearance represented 9.5% of g.f.r. These data are incompatible with the current theory that urate only appears in human urine when a tubular reabsorptive mechanism of limited capacity is exceeded. An alternative, but improbable, hypothesis is that under normal conditions tubular reabsorptive mechanisms are not saturated but do not act rapidly enough to extract urate completely from the glomerular filtrate. From consideration of certain pharmacological data, and from the fact that only about 6% of the plasma urate passes into the C.S.F. (J. Clin. Invest. **26:** 991, 1947), it appears probable that only a small fraction of the

plasma urate is ultrafiltrable through the human glomerulus. As only 20% to 30% of the plasma urate is bound to plasma protein in ultrafiltration experiments, the failure of most of the plasma urate to pass the human glomerulus appears not to result from protein binding. An alternative possibility is that the major portion of the plasma urate circulates as polymeric urate complexes. This state has been demonstrated to occur in the azotemic chicken. (Am. J. Physiol. *in press.*)

Anaphylactic shock in a restricted circulation system excluding the liver in the dog. PARKE H. WOODARD (introduced by K. E. JOCHIM). *Dept. of Physiology, Univ. of Kansas, Lawrence.* Most previous work has supported the contention that it is necessary for the liver to remain in the circulation for the drop in blood pressure to occur in canine anaphylaxis. However, some recent work in other laboratories presents evidence contrary to this view. The experiments here presented indicate that not only is the liver unnecessary but that the fall in pressure occurs in a somewhat restricted circulation system.

Dogs were sensitized to horse serum and after the lapse of 12 to 14 days each animal was tested for sensitivity by determining the ability of its blood to passively sensitize normal dogs. If proven sensitive, each dog was anesthetized with nembutal intravenously and after opening the chest, the following vessels were ligated: the azygos, internal mammary and right subclavian veins, the right subclavian and internal mammary arteries, the vena cava above the diaphragm and between the liver and the diaphragm, and the aorta just distal to the left subcalvian artery. Blood pressure was recorded from the left subclavian artery.

When a shock dose of horse serum was introduced intravenously into this preparation, the characteristic fall in blood pressure resulted. In the animals that were tested there was no change in blood coagulation time.

Photoelectric determination of blood oxygen saturation in man. E. H. WOOD, J. E. GERACI (by invitation) and D. L. GROOM (by invitation). *Section on Physiology of the Mayo Clinic and Foundation, Rochester, Minnesota.* The Millikan oximeter earpiece has been modified so that a transparent pneumatic pressure capsule is interposed between the light source and the ear. This earpiece makes possible measurement of the red and infra-red light transmission of the bloodless (pressurized) ear and of the normal heat flushed ear. The differences in light transmission between the bloodless and flushed ear are a measure of red and infra-red transmission of the blood interposed in the optical path of the earpiece. The ratio of the light transmission of blood in the red and infra-red is a function of the oxygen saturation of this blood. The relationship between this ratio and arterial oxygen saturation has been determined by comparing the results of Van Slyke analyses of 143 arterial blood samples from 37 individuals with the average results of instrumental readings obtained during the period of withdrawal of the samples. Normal white and negro subjects were used and patients with suspected arterial hypoxemia. Arterial samples were obtained during rest, exercise and while breathing gas mixtures of from 100 to 8 per cent oxygen content. The standard deviation of the differences of the individual photoelectric determinations from the mean calibration line was 2.4 percentage saturation points. On the same basis a photoelectric cuvette has been constructed in which the blood is contained in a 2 mm. I.D. polythene tube (volume 0.5 cc.). This device attached to a cardiac catheter has been used for immediate determination of the oxygen saturation of cardiac blood samples during diagnostic cardiac catheterizations.

A technique for obtaining multiple arterial blood samples applied to the study of cyanosis in man. (Kodachrome Motion Picture). E. H. WOOD, G. E. MONTGOMERY, JR. (by invitation) and J. E. GERACI (by invitation). *Section on Physiology of the Mayo Clinic and Foundation, Rochester, Minnesota.* The sampling system which is coated internally with silicone and has a volume of 0.6 cc. consists of: an internally polished No. 20 needle, fitted into a glass adapter which is connected to a metal stopcock via a 15 centimeter length of 2 mm. I.D. polythene tubing. The stopcock is adapted for connection to a hypodermic syringe. The syringe and sampling system are filled with sterile isotonic saline containing 20 mgm. of heparin per liter. To facilitate the arterial puncture an armboard is used which fixes the hand in the position of extreme extension at the wrist. The site of the puncture (radial artery near the styloid process) is infiltrated with 1.0–2.0 cc. of 2 per cent procaine hydrochloride. After the puncture is completed and the needle taped in position, arterial samples may be obtained at will by opening the stopcock. Aproximately 1 cc. of blood is discarded before beginning collection of each sample. The system is cleared of blood after each sampling period by reinjection of a few cubic centimeters of the sterile heparinized isotonic saline solution. This procedure has been carried out on 90 individuals. Approximately 500 arterial samples have been obtained from these subjects while at rest, breathing various oxygen mixtures, or walking on a power driven treadmill. The technique has been used to study the correlation of arterial oxygen saturation with clinical estimations of the degree of cyanosis induced in normal negro and white subjects by breathing low oxygen mixtures.

The pattern of cutaneous representation in the rat's cerebral cortex. CLINTON N. WOOLSEY and

D. H. LeMessurier (by invitation). *Dept. of Physiology, Johns Hopkins University, School of Medicine, Baltimore 5, Md.* Somatic areas I and II were mapped in the rat's cortex by the evoked potential method. Of particular interest is the simplicity of the pattern of representation in somatic area I (postcentral homologue). The pattern may be visualized by imagining a rat lying, right side up, on the left hemisphere with its tail near the midline and with the mid-dorsal line of the body from tail to nose extending across the hemisphere along the rostral border of the visual, auditory and somatic II areas. The leg, arm and face representations then, extend rostralward in the cortex so that the pattern presented is a somewhat distorted picture of a rat with its various parts related to one another in much the same way as in the actual animal. The results call for a re-examination of the details of tactile representation in other species so that the evolution of the pattern as seen in primates may be understood.

Relation of retinal and optic nerve response to intensity of illumination of the grasshopper eye. Verner John Wulff (by invitation) and Theodore Louis Jahn. *Department of Zoology and Physiology, University of Illinois, and Departments of Physiology and Zoology, State University of Iowa.* Illumination of the eye of grasshoppers (Melanoplus differentialis) results in electrical responses in the visual system that can be recorded in the following temporal order: 1) the retinal action potential and 2) a slow negative variation on the surface of the nerve between the optic ganglion and the brain. The latter potential is presumably a sign of the asynchronous activity of neurones within the optic ganglion. The latency of these responses, measured from the beginning of the light stimulus, increases as the stimulating light intensity decreases. These relations are such that the time interval between the beginning of the retinal response and the beginning of the nerve response called the retinal-nerve interval also increases as the intensity of stimulation decreases. This regular increase in the retinal-nerve interval is correlated with a reduction in magnitude of the retinal action potential and an increasing delay in the time of occurrence of the maximum of this action potential. In other words, the retinal-nerve interval increases as the rate of change of potential across the retina decreases. This increase in the retinal-nerve interval supports the hypothesis that the retinal action potential is the process or a sign of the process which is responsible for the activation of nervous elements in the optic pathway.

The effect of local graded pressure upon electrical activity and excitability of the motor cortex. J. E. Ziegler and T. W. Rasmussen (introduced by Herbert Jasper). *Department of Neurology and Neurosurgery, McGill University and the Montreal Neurological Institute.* The electrical activity and excitability of the motor cortex was measured beneath a disc one half to 2 cm.², during the application of pressures between 20 and 70 grams. At a critical pressure per unit area, the electrical activity and excitability of the cortex beneath the disc disappeared without significant change in the cortex adjacent to the disc or from other portions of the ipselateral or contralateral hemisphere. With a 0.5 cm.² disc, the cortical excitability was suppressed within 30 to 60 seconds following the application of 30 grams of pressure. With a 1 cm.² disc, the electrical excitability disappeared within 1 to 12 minutes (average 3.6 min.) at 50 to 60 grams pressure, and between 1 and 5 minutes (average 2.8 min.) with 70 grams pressure. The local electrical activity from bipolar recording electrodes beneath the 1 cm.² disc disappeared with 70 grams of pressure for 1 to 5 minutes (average 2.6 min.). Local electrical activity tends to disappear in about the same time as does electrical excitability although several exceptions occurred with pressures of 50 to 60 gram. The suppression of cortical fuction was confined to the pressure area and was completely reversible in most instances and did not result in significant pathological changes in the affected cortex. If slow waves were present in the electrical activity due to local injury of the cortex, these waves disappeared earlier than did the bursts of rhythmic waves, characteristic of the animal under nembutal anaesthesia. Further experiments showing the use of this method in the differential analysis of various components of the electrocorticogram will be described.

The effect of thrombin injections on hemostasis. Marjorie B. Zucker. *Department of Physiology College of Physicians and Surgeons, Columbia University.* In an attempt to study hemostasis in fibrinogenopenic dogs under nembutal, 2000 to 4000 units of Parke-Davis thrombin (125 units/cc.) were given through a catheter inserted in the jugular vein (0.18 cc./min.) without untoward effects. After injection, blood samples showed less than 10 mg. per cent fibrinogen, between 10 and 20% prothrombin activity (Quick method with addition of fibrinogen, calculated from dilution curve), gross hemolysis, normal clot retraction, no fibrinolysis, and a reduction in platelets which however, fell below 150,000 mm. in only one of seven experiments. In two experiments the two-stage prothrombin method was also used, and showed only a 30% depression of prothrombin. The one-stage prothrombin times were increased less by addition of an equal volume of prothrombin-free plasma than by oxalated saline. These data suggest that there is a deficiency of the Ac factor as well as of prothrombin.

Bleeding times from nicked ear veins were greater than 10 minutes and usually no hemostasis oc-

curred. Glass A-V shunts were not occluded by platelet thrombi although a thin coating frequently formed inside the cannulae. Studies of successive smears from a venous blood sample (Pinniger and Prunty, Brit. J. Exp. Path., 27: 200, 1946) showed smaller groups of platelets and sometimes delayed and diminished metamorphoses. Autopsy failed to disclose microscopic clots or evidence of purpura.

Administration of fibrinogen failed to influence the bleeding time. Whole blood infusion reduced the bleeding time to normal and was followed by occlusion of the shunt.

Hepato-renal factors in circulatory homeostasis. XVII. Relation of renal VEM to vascular compensation in shock. B. W. ZWEIFACH, S. BAEZ (by invitation), R. F. FURCHGOTT (by invitation) and EPHRAIM SHORR. *Dept. of Medicine, Cornell University Medical College and The New York Hospital, New York City.* There regularly appears in the blood during the compensatory stage of shock a vaso-excitor principle (VEM) of renal origin which produces in normal test rats hyper-reactive effects on the terminal vascular bed similar to those observed in the same vessels of the shocked animal. Experiments were designed to determine to what extent the compensatory reactions of the peripheral blood vessels during shock are dependent upon participation of the renal VEM mechanism. Anesthetized dogs, subjected to hemorrhage following occlusion of the renal circulation, had no VEM in their blood and developed an accentuated vaso-depressor reaction. VEM was likewise absent from the blood of arenal animals subjected to lethal tourniquet shock and VDM, from the damaged skeletal muscles, appeared in the blood within 10 minutes after tourniquet release. Whereas, in animals with intact kidneys, this VDM is masked in the blood by the preponderance of the oppositely-acting renal VEM. The relation of the kidney to the compensatory vascular response to shock was further emphasized by experiments in unanesthetized arenal dogs subjected to a type of hemorrhagic shock from which unanesthetized dogs with intact kidneys are recoverable by transfusion. In the unanesthetized arenal animal, the initial hyper-reactive stage was absent and hypo-reactivity developed with unusual rapidity. These animals failed to respond to replacement of the blood withdrawn. Microscopic observation of the omental circulation was also used to determine the effect of the exclusion of the kidney on the capacity of the animal to establish adequate compensatory vasoconstrictor changes in the peripheral blood vessels during shock.

Hepato-renal factors in circulatory homeostasis. XVI. Vascular changes in mesentery in renal hypertension in rats. B. W. ZWEIFACH, S. ROSENFELD (by invitation) and EPHRAIM SHORR. (*Dept. of Medicine, Cornell University Medical College and The New York Hospital, New York City*). A study was made of the changes in the responses of the terminal arterioles and precapillaries in the mesoappendx during the development of hypertension in the rat following the application of a gauze-collodion sac about the kidneys. In control animals the reactivity of the terminal blood vessels to topical epinephrine remains relatively constant, the minimal effective concentration of epinephrine required to produce a slowing of the capillary circulation usually ranging between 1:2 to 1:4 million. Within 2 weeks following the application of a gauze-collodion sac the response to epinephrine becomes enhanced, the threshold concentrations being as low as 1:8 to 1:32 million. Not only is the response to topical epinephrine enhanced but the reaction to intravenously administered VEM is also potentiated. A significant finding during the development of hypertension was the marked increase in the number of capillary vessels in the mesentery which appeared within 2 to 3 weeks after application of the sac to the kidney. Initially all the vessels show an overall rapid circulation. In a later stage a rapid flow persists only through thoroughfare channels, which have a varicose appearance with prominent, thickened, smooth muscle cells. The precapillary sphincters are narrowed so that the capillary flow is sporadic. Little or no vasomotion is seen in the precapillary vessels. The flow in the collecting venules is unusually rapid. These vascular phenomena closely resemble those prevailing during the initial compensatory stage of experimental shock, when VEM predominates in the blood stream.

THE AMERICAN SOCIETY OF BIOLOGICAL CHEMISTS, INC.

THIRTY-NINTH ANNUAL MEETING

Atlantic City, New Jersey, March 15, 16, 17, 18, 19, 1948

(For possible corrections in any of the following abstracts see the next issue.)

Fractionation studies on the cytoplasmic proteins of human lymphoid tissue. ADOLPH ABRAMS (by invitation) and PHILIP P. COHEN. *Department of Physiological Chemistry, University of Wisconsin Medical School, Madison, Wisconsin.* A technique for obtaining reproducible electrophoretic patterns of soluble proteins in extracts from human lymphoid tissue (tonsil) has been established. Although complete resolution of all the components of the system is not realized, definite groups of proteins can be identified by a characteristic mobility corresponding to the maximum ordinate of well defined peaks in the pattern. The complex mixture was fractionated with 30, 55, and 100 per cent saturation of $(NH_4)_2SO_4$, and each fraction was analyzed electrophroetically. Each group of proteins had reproducible solubility properties under the conditions used. A comparison of the solubility properties and mobilities of the tissue proteins with those of serum has allowed a tentative conclusion as to the possible relationship between certain serum proteins and the proteins of lymphoid tissue.

Small amounts of a fast component (mobility about 12×10^{-5} cm²/volt/sec.) have been observed consistently. Chemical and spectrophotometric analysis indicate that this fast component is free ribonucleic acid. A fraction of lymphoid tissue prepared by high speed centrifugation, corresponding to the "microsome" fraction was found to have a composition, as to nitrogen and phsophorus, similar to "microsomes" prepared by other workers from different tissues. The ribonucleic acid content of this fraction was of the order of 4 per cent.

A micromodification of the Folin method for estimating urinary creatinine. ROBERTO REYNAUD ACOSTA (by invitation) and ROBERT E. JOHNSON. *U. S. Army Medical Nutrition Laboratory, 1849 W. Pershing Road, Chicago 9, Illinois.* A micromodification of the Folin method for estimating urinary creatinine has been developed for the Coleman Junior Spectrophotometer, Model 6, with #6-310 cuvettes. All reactions are carried out in the cuvette. The only reagent is an aqueous solution containing 0.1% picric acid and 1% sodium hydroxide. Steps in the procedure are: pipette 0.05 ml. urine into cuvette, add 10 ml. of alkaline picrate solution, stand 15 minutes, read, wave length 520. Study was made of the following factors: initial volume of water; concentration of picric acid; concentration of alkali; mixing of picric acid and alkali; time of standing before reading; temperature of reagents; stability of color. Statistical comparison was made between the modified and the original Folin methods in relation to creatinine standards, urine and urine to which creatinine was added.

The penicillins produced by Aspergillus flavus. M. ADLER (by invitation) and O. WINTERSTEINER. *Division of Organic Chemistry, Squibb Institute for Medical Research, New Brunswick, N. J.* Evidence obtained in an earlier investigation (Fried, Koerber and Wintersteiner, J. Biol. Chem., **103,** 341 (1946)) suggested that "flavacidin," the penicillin-like antibiotic produced by *Aspergillus flavus* in submerged culture, consisted chiefly of 3-pentenylpenicillin, $CH_3 \cdot CH{=}CH \cdot CH_2 \cdot CH_2 \cdot C_9H_{11}O_4N_2S$, a double bond isomer of the 2-pentenylpenicillin (F - penicillin, $CH_4 \cdot CH_2 \cdot CH{=}CH \cdot CH_2 \cdot C_9H_{11}O_4N_2S$) elaborated by *Penicillium notatum.* This conclusion was based on the properties of the penilloaldehyde dinitrophenylhydrazone, particularly on the identity of its X-ray diffraction pattern with that of the dinitrophenylhydrazone of synthetic 4-hexenoylaminoacetaldehyde, $CH_3 \cdot CH{=}CH \cdot CH_2 \cdot CH_2 \cdot CO \cdot NH \cdot CH_2 \cdot CHO$. In contrast, a recently prepared batch of "flavacidin" was found to contain benzylpenicillin (G-penicillin) as the main component. The remainder of the active material consisted largely of *n*-amylpenicillin (dihydro F-penicillin), an entity known to be produced also by *Aspergillus giganteus* and by *Penicillium notatum.* In this case the R-group present was unequivocally identified by degradation to the acid $R \cdot COOH$, *n*-caproic acid. It was furthermore ascertained that the dinitrophenylhydrazone of *n*-caproylaminoacetaldehyde (and of the penilloaldehyde obtained from the *n*-amylpenicillin fraction) cannot be differentiated either by melting point or X-ray data from that of 4-hexenoylaminoacetaldehyde. Thus the original conclusion as to the occurrence of a 3-pentenylpenicillin in "flavacidin" has been deprived of most of its basis. The search for an unsaturated (F-type) penicillin in the present material resulted eventually in the isolation of small amounts of an oxidation product, propionaldehyde, which can be derived only from 2-pentenylpenicillin.

Detoxification of diphtheria toxin by peroxidase. KJELL AGNER (Introduced by A. M. PAPPENHEIMER, JR.). *Chemical Laboratory of the Serafimer Hospital, Stockholm, Sweden, and the Department of Bacteriology, New York University College of Medicine.* Previous experiments have shown that both crystalline horse radish peroxidase and purified verdoperoxidase from leucocytes, in the pres-

ence of 0.001 M hydrogen peroxide, detoxify crude diphtheria toxin produced on a peptone-containing medium. It has now been demonstrated that toxin produced on the chemically defined medium of Mueller and Miller is also detoxified by peroxidase. The detoxified material still flocculates with antitoxin and, therefore, has the properties of a toxoid. Highly purified diphtheria toxin was not appreciably detoxified by 0.001 M hydrogen peroxide in the presence of peroxidase. The reaction, therefore, appears to require the presence of some intermediate substance present in crude culture filtrates.

The sulfur amino acid requirement of the infant. ANTHONY A. ALBANESE, L. E. HOLT, JR., VIRGINIA I. DAVIS (by invitation), SELMA E. SNYDERMAN (by invitation), MARILYN LEIN (by invitation) and EMILIE M. SMETAK (by invitation). *New York University College of Medicine.* The cystine and methionine requirements of the infant were determined by measuring the nitrogen retention and weight gains on a diet prepared by using a diet deficient in these 2 amino acids as the principal source of nitrogen. This diet was supplemented with varying proportions of L-cystine and L-methionine. Five male infants (4–11 months) were studied. In 3 infants who received no cystine supplements the nitrogen retention and body weight gains were restored to normal levels by an intake of 99 mg. of L-methionine per kilo of body weight. In 2 infants who received a 1 per cent L-cystine supplement the nitrogen retention and positive weight change were restored to normal values by an intake of 79 mg. of L-methionine per kilo of body weight.

These findings suggest that cystine is capable of stimulating growth only when methionine is present in suboptimal amounts and that about 20% of the methionine needs can be met by cystine. Moreover, it appears from these results that the quantities of sulfur amino acids provided by the commonly employed infant formulae prepared from cow's milk are adequate. On the basis of calculations from the available data on the sulfur amino acid content of human milk, it would appear that the breast milk provides a very meagre margin of safety during the early months of life and that breast milk does not provide an adequate quantity of the sulfur amino acids in the latter part of the first year. The margin of safety is considerably greater in the artificially fed infant.

Adenine-pentose-pyrophosphate from mung beans. H. G. ALBAUM, M. OGUR (by invitation), H. MENDELOW (by invitation), and A. HIRSHFELD (by invitation). *Departments of Biology and Chemistry of Brooklyn College, Brooklyn, N. Y.* When a trichloracetic acid filtrate from fresh mung bean sprouts is neutralized and treated with barium acetate, a precipitate is obtained which, in addition to large amounts of inorganic phosphorus, contains adenine, pentose, and labile phosphorus. Application of the Needham procedure for the isolation of adenosine triphosphate from this precipitate has yielded unsatisfactory results in our laboratory. If instead, the barium precipitate is suspended in 0.1 M acetate buffer, pH 4.35, and centrifuged, the supernate contains more than 90% of the inorganic phosphorus and about 30% of the adenine. Treatment of the residue with excess sodium sulfate yields a supernate which contains almost all of the remaining adenine. The ratio of adenine:pentose:labile phosphorus:total phosphorus is approximately 1:1:2:3. When this supernate is treated with silver nitrate a precipitate is obtained which, on decomposition with hydrogen sulfide and treatment with barium acetate, yields a barium insoluble fraction with a molar ratio of adenine:pentose:labile phosphorus:total phosphorus of 1:1:2:3. All of the labile phosphorus is split by the potato pyrophosphatase and half is donated to glucose in the yeast hexokinase system at a rate comparable to animal adenosine triphosphate.

Repletion of protein depleted dogs with whole egg and wheat gluten proteins. JAMES B. ALLISON, JOHN A. ANDERSON (by invitation), and JOHN I. WHITE (by invitation). *Bureau of Biological Research, Rutgers University, New Brunswick, New Jersey.* Dogs were depleted in protein by feeding a protein-free diet. They were repleted for 30 days by feeding a defatted, dried, whole egg protein or by feeding wheat gluten. The nitrogen balance indexes of the whole egg protein increased above control values in the depleted dog returning to control values upon repletion. Similarly, the index for wheat gluten was greater than control values when the protein was fed to depleted dogs. The excretion of urinary nitrogen on a protein-free diet, (UNo) which reflects the magnitude of the protein stores, decreased upon depletion and returned toward control values with repletion on whole egg protein. Repletion with wheat gluten, however, decreased the UNo even below values obtained during the protein depleted state. Thus, feeding wheat gluten nitrogen did not increase, rather decreased, those protein stores which are involved in the formation of minimum catabolic nitrogen. These results may be interpreted to mean that body protein stores may be utilized to supplement the wheat gluten, an interpretation which is supported by data obtained in normal dogs. Some tissue stores were repleted with the wheat gluten, however. The body nitrogen gain and the plasma protein nitrogen gain with wheat gluten was equal to approximately half that obtained with whole egg which increased body nitrogen and plasma protein nitrogen almost to control values.

The rate of disappearance of human albumin in normal and protein depleted dogs. CARL ALPER

(by invitation) SHIRLEY DEBIASE (by invitation) and BACON F. CHOW. *Division of Protein Chemistry, The Squibb Institute for Medical Research, New Brunswick, New Jersey.* The study of the rate of disappearance of a foreign protein from the circulation, although of fundamental importance in protein metabolism, has been neglected generally because suitable quantitative techniques for the estimation of the foreign protein were not available. Recently, an immunological method was described for the quantitative determination of human albumin. This method is satisfactory for the estimation of human albumin in the presence of other albumins. Normal dogs and dogs depleted by plasmapheresis were injected with approximately 250 mgm. of normal human serum albumin per kgm. body weight. Samples of blood were removed at fixed time intervals for 96 hours and the amount of serum albumin in the circulation was derermined by the immunological method and by the determination of total plasma volume. The serum albumin is tolerated very well by the normal and depleted dogs. There is no significant change in plasma volume after the injection of serum albumin. The disappearance of serum albumin is rapid in the first 24 hours and then the rate gradually becomes asymptotic. Approximately one-third of the infused albumin disappears in 24 hours; and an additional one-third disappears in the next 72 hours.

Aminoaciduria in progressive muscular dystrophy. STANLEY R. AMES and HUGH A. RISLEY (introduced by P. L. HARRIS). *Laboratories of Distillation Products, Inc., Rochester, New York.* Wasting of muscle is characteristic of progressive muscular dystrophy but no significant changes in total nitrogen excretion are observed. It seemed probable that there might be an increased urinary excretion of minor nitrogenous products in addition to creatine, such as free amino acids. The use of partition chromatography to determine the urinary distribution and excretion of amino acids in humans was developed by Dent (Lancet, II, 637 (1946)). He reported low concentrations of two and occasionally three or four amino acids in urine of normal humans. A total of 80 urine samples were obtained from patients who had been diagnosed as suffering from progressive muscular dystrophy. The urines were chromatographed on filter paper employing the "strip" technique with the following solvents saturated with water: butanol, phenol, and lutidine-collidine (50-50). The results of the "strip" chromatograms indicated that practically all urines from patients not receiving therapy were markedly increased in amino acid excretion as compared with normals. Two-dimensional partition chromatography was also employed using phenol and lutidine-collidine (50-50) as solvents. Compared with a normal picture of from 2 to 4 spots on the chromatogram, the majority of urines from patients not receiving therapy showed 8 to 11 or more spots. As many as 15 separate spots have been observed. On the basis of these urine specimens, it can be concluded that the condition of human progressive muscular dystrophy is characterized by a generalized aminoaciduria.

The estimation of functional hepatic mass in normal and phosphorus poisoned dogs. CARL E. ANDERSON (by invitation), ROBERT G. GALE (by invitation) and C. S. ROBINSON. *Departments of Biochemistry and Medicine, Vanderbilt University, Nashville, Tennessee.* Mason has reported (Federation Meeting, May 1947) a constant injection technique for estimating the functional hepatic excretory mass for dogs based on the principle that the maximum rate of hepatic excretion of bromsulphalein (Lm) is proportional to the functional hepatic mass. His values for Lm ranged from 7.5 to 10.3 mg./min./sq. meter surface area, and averaged 0.45 mg./kg. body wt./min. We have obtained values from 5 normal dogs (15 experiments) of average weight (10–13 kgs.) which confirm Mason's constants.

Dog	Lm	
	Mgs./kg./min.	Mgs./min./sq. m. S. A.
1	0.48	9.5
2	0.47	9.1
3	0.56	10.8
4	0.42	8.3
5	0.42	8.9

Two dogs poisoned by the daily subcutaneous injection of phosphorus (0.4 mg./kg./P_4) showed reduction in Lm. Dog *2* died when a value of 5.8 mg./min./sq.m.S.A. (0.28 mg./kg./min.) was obtained. Gross examination of the liver revealed two areas of hemorrhagic degeneration in the parenchyma. Severe liver necrosis was observed histologically. Dog *5* was sacrificed when the value for Lm reached 6.7 mg./min./sq.m.S.A. (0.31 mg./kg./min.). Histological examination of the liver revealed only a slight cloudy swelling. The prothrombin time remained essentially normal. Present observations indicate that values representing a functional change in the liver without apparent histological change may be obtained.

On the fate of labeled pyruvic acid in the intact animal. H. S. ANKER (introduced by E. A. EVANS, JR.). *Department of Biochemistry, University of Chicago.* Pyruvic acid labeled with C^{14} at carbon atom 2 was synthesized from C^{14} carbon dioxide via acetic acid, pyruvonitrile and pyruvic acid amide. Pyruvic acid was injected intraperitoneally over a period of 3 days into a rat which was kept on a high protein diet. p-Amino benzoic acid was fed simultaneously. The following compounds were isolated

and their C^{14} content determined. They are given in the order of decreasihg radioactivity. Urea, acetyl p-amino benzoic acid, liver protein, glycogen, saturated liver fatty acids, liver cholesterol, saturated carcass fatty acids, carcass cholesterol and hemin. From the radioactivity of the isolated compounds it can be concluded that the carbon atoms of pyruvic acid are utilized in the synthesis of all the compounds mentioned. The bulk of the recovered C^{14} was found in the fatty acids.

Partial hydrolysis products of human hair. JAMES C. ANDREWS. *Department of Biological Chemistry and Nutrition, School of Medicine, University of North Carolina, Chapel Hill, N. C.* Slow hydrolysis of human hair by means of 3 M H_2SO_4 at 38°C. produces a solution of proteose-peptone corresponding in amount to about half the original weight of the hair. The protein in the solution has a free amino to total nitrogen ratio of about 39%, a cystine sulfur content of 2.90% and an optical activity of −50. Variations in the time of hydrolysis from 4 months to over 4 years yield a product in which the above figures remain very constant. The total non-sulfate sulfur of these solutions is about twice the amount of cystine sulfur. The evidence indicates that the non-sulfate, non-cystine sulfur is partly, if not all, in the form of cysteic acid. This latter has evidently resulted from atmospheric oxidation of cystine produced in amounts too large to find place in the structure of the proteose and the peptone. The insoluble residue from such hydrolyses still retains the gross and microscopic appearance of the original hair. However, it is highly brittle and easily crumbles to a powder. It contains about 12% nitrogen and 5.2% sulfur but produces no free cystine on complete hydrolysis.

Fractionation of urinary steroids by use of the Craig counter current distribution machine. REGINALD M. ARCHIBALD and E. STROH (by invitation). *Department of Biochemistry, School of Hygiene and Public Health, Johns Hopkins University, Baltimore, Maryland.* Carbon tetrachloride extracts of hydrolized, human urines from normal individuals and from those with disturbed production or metabolism of hormones of the suprarenal glands or gonads have been fractionated in the Craig machine using equal volumes of cyclohexane and a 38% (by volume) solution of alcohol in water. Analysis of fractions by use of a) spectrophotometric determination of the absorption of ultra violet light, b) photofluorometric measurement of the fluorescence produced on heating fractions with 90% H_2SO_4, c) Zimmermann's reaction with alkaline m-dinitrobenzene showed the presence of 8 or more components. Attempts are being made to effect further fractionation and identification of the components. The fluorescence resulting from heating fractions with 90% H_2SO_4 paralleled the absorption of light of $\lambda = 410$ mu by these heated fractions.

Reaction of plasmochin with formaldehyde. R. M. ARCHIBALD and JAMES R. WEISIGER (by invitation). *Hospital of The Rockefeller Institute for Medical Research, New York.* An understanding of the degradation of plasmochin *in vivo* is basic to an understanding of its pharmacological action. It reacts readily in aqueous formaldehyde solution binding irreversibly one equivalent of acid and one molecule of formaldehyde. The structure of the strongly fluorescent quaternary salt which can not couple with diazoreagents and which is 20 times more toxic than plasmochin was confirmed by analysis of the chloroplatinate.

$C_{20}H_{31}N_3OPtCl_6$

Calc. C: 32.59, H: 4.23 N: 5.69.
Found C: 32.28, H: 4.16 N: 5.67.

This compound decomposes irreversibly in alkaline solution to give at least two new compounds, neither of which is soluble in non-polar solvents, suggesting the persistence of the quaternary structure. These properties have prevented the recovery of any of the products from admixture with biological material. A comparable *in vivo* reaction product would not be detected by the ordinary methods of isolation and analysis, in accord with the observed rapid disappearance of plasmochin from the blood. The toxicity of the drug to parasite or to host may result from reaction with a naturally occurring aldehyde or ketone.

Tissue incorporation and excretion of radioactive carbon administered as carbonate. W. D. ARMSTRONG, JACK SCHUBERT (by invitation), and ARTHUR LINDENBAUM (by invitation). *Department of Physiological Chemistry, University of Minnesota Medical School, Minneapolis, Minnesota.* The excretion of C^{14} in expired air, urine and feces by mature rats has been determined over periods of 6–40 days after intraperitoneal injection of $Na_2C^{14}O_3$ and after intraperitoneal implantation of $CaC^{14}O_3$ as a powder or as pellets. The absorption of C^{14} from pellets of $CaC^{14}O_3$ is slow and allows a greater incorporation of the isotope in organic compounds and in the bone salt. In 40 days all but 0.38 ± 0.053% of C^{14} administered as powdered $CaC^{14}O_3$ is excreted. These data and the results indicating the fraction of the total dose of C^{14} found in the skeleton, enamel, dentine, bone inorganic carbon, bone protein, liver and muscle glycogen, glycerol, fatty acids, muscle protein, hemin and in other tissues and compouds will be presented.

The action of ethanolamine, monomethyl- and dimethyl-ethanolamine on lipide phosphorylation. CAMILLO ARTOM and W. E. CORNATZER (by invitation). *Department of Biochemistry, Bowman Gray School of Medicine, Wake Forest College, Winston-Salem, North Carolina.* Rats maintained for 7 days on a low fat, low protein diet received by

stomach tube a single dose (0.2 millimoles in 1 cc. of water) of ethanolamine, monomethyl-, dimethyl-ethanolamine, or choline. Controls received 1 cc. of water. The rats were then injected with P^{32} (as Na_2HPO_4). After 6 hours the lipides were extracted from the liver and small intestine and their radioactivity and phosphorus content were determined. All substances tested stimulated lipide phosphorylation in both tissues. After giving ethanolamine, monomethyl-, dimethyl-ethanolamine, and choline, the average increases in the specific activity above the controls were 43, 65, 56 and 21 per cent in the liver, and 13, 20, 36 and 16 per cent in the intestine, respectively.

In a few experiments the separation of the liver phospholipides into choline-containing (C.c.) and non choline-containing (N.c.c.) was attempted (see Taurog *et al.*, J. Biol. Chem., **155**, 19 (1944)). In almost all experiments increases in the radioactivity of both fractions were observed. However, the increase was proportionately greater in the N.c.c. fraction after giving ethanolamine. It was approximately the same in both portions after feeding monomethyl-ethanolamine. On the other hand, it was definitely greater in the C.c. fraction when dimethyl-ethanolamine and especially when choline had been administered.

On the absorption of phospholipides. CAMILLO ARTOM and MARJORIE A. SWANSON (by invitation). *Department of Biochemistry, Bowman Gray School of Medicine, Wake Forest College, Winston-Salem, North Carolina.* Labelled phospholipides (obtained from the livers of rats injected with radioactive phosphate) were fed by stomach tube to fasting rats. After 3 or 6 hours the radioactivity and phosphorus content were determined in the inorganic and lipide fractions of plasma and liver. The results were compared with those obtained on rats receiving by stomach tube non-labelled phospholipides and P^{32} as sodium phosphate. In these controls, at both time intervals, the specific activities were in the following order: liver inorganic P > plasma inorganic P > liver lipide P ≧ plasma lipide P. These findings are consistent with the assumption that the phospholipides synthesized in the liver from the absorbed inorganic phosphate were the main or only source of the labelled lipides in plasma. On the other hand, in all experiments in which radioactive phospholipides were fed, the gradient of the specific activities was the following: plasma lipide P ≧ plasma inorganic P ≧ liver inorganic P > liver lipide P. The presence after 3 hours of large amounts of isotopic phosphorus in the inorganic fraction of the liver and especially in that of the plasma indicates that the phosphate was rapidly split from the lipide in the intestinal tract and absorbed. However, the high specific activity of the phospholipides in plasma is definite evidence that they cannot derive solely from the liver lipides, but that a detectable amount of the ingested phospholipides has also been absorbed as the intact molecule.

Folic acid requirement of the rat and some characteristic lesions observed in the deficient animals. CONRADO F. ASENJO. *Department of Chemistry and Nutrition, School of Tropical Medicine, San Juan, Puerto Rico.* White rats (Wistar Institute strain), fed since their 21st. day a highly purified diet, high in carbohydrate and containing 2% succinylsulfathiazole, besides all the known nutritional requirements except folic acid, showed cessation of growth, leukopenia, and the characteristic skin lesions of folic acid deficiency in an average of 4 to 5 weeks. These animals were supplemented at this time with different levels of folic acid administered 6 times per week for a period of 3 weeks. One group was left as negative controls. Gain in weight, leukocyte response, and per cent survival were recorded.

According to these results, the minimum amount of folic acid required by these strains of rats for maintenance is in the neighborhood of 0.5 μg. per day six times per week. However, this amount of folic acid does not improve the white cell count during the supplementation period. To make appreciable gains in weight and to recover the normal white cell count in the 3 week period these rats seem to require somewhere between 1–8 μg. per day. All negative controls, as well as 33% of animals receiving 0.5 μg. of folic acid, were found at autopsy to have developed infarcts in the spleen. None of the animals receiving 1 μg., or more, of folic acid per day developed this lesion. The immediate cause of these infarcts, according to Dr. Enrique Koppisch, of our Department of Pathology, seems to be a thrombus formation in the splenic vein.

Distribution pattern of carbonic anhydrase in the fetal central nervous system. WINIFRED ASHBY (introduced by DR. JOSEPH H. ROE). *Blackburn Laboratory, St. Elizabeths Hospital, Washington.* The quantitative distribution of carbonic anhydrase in the central nervous system has been studied in 7 prematurely born human infants and one full term, and in 5 fetuses, obtained from cattle slaughtered for beef, ranging in age from 3 to 7½ months. As distinguished from the adult pattern of distribution, carbonic anhydrase was not found in the cerebrum of the fetus, although it occurred in measurable and in progressively increasing amounts in phylogenetically older parts of the central nervous system. In the human fetus the comparison between the content of the medulla and the pons, which in the adult gave an excess in the pons, showed an excess in the medulla in the fetus. In two instances in which the cord was obtained a markedly greater content was found in the cord than in any other part studied as contrasted with the adult in which higher centers

greatly exceeded the cord in carbonic anhydrase content. That the lack of carbonic anhydrase content in the cerebrum of the premature human infants, which did not survive, was not abnormal, would seem to be supported by the negative findings in the cerebrum of the cattle fetus examined up to 7½ months of fetal life. In a calf a few days after birth a cerebral content of 7.8 units was found as compared with 13.5 units in the hemispheres of the mature animal.

Effect of carbon dioxide concentration on brain lactate. JAMES A. BAIN (by invitation) and J. RAYMOND KLEIN. *Departments of Pharmacology, Psychiatry, and Biological Chemistry, University of Illinois College of Medicine, Illinois Neuropsychiatric Institute, Chicago.* In cats, it was found that the following experimental conditions: 1) breathing mixtures containing 5-30% carbon dioxide and 95-70% oxygen, 2) breathing carbon dioxide mixtures during intravenous administration of sufficient sodium bicarbonate to maintain the plasma pH at a control level, and 3) breathing oxygen during administration of bicarbonate result in concentrations of brain lactate that are lower than the level found in cats breathing room air. Compared to the brain lactate level found in air, the experimental conditions produced decreases in level of about 60, 30, and 10% respectively. The results suggest that the effect of carbon dioxide is not due to change in plasma hydrogen ion concentration alone, but depends also upon the concentration of total carbon dioxide, *i.e.* carbon dioxide plus carbonic acid. It is not clear as yet whether known effects of increased carbon dioxide concentration, *i.e.* increase in cerebral blood flow and consequently in oxygen tension, account completely for the changes in brain lactate or whether other factors are involved.

Circulatory disturbances in hepatic and renal cortical necrosis. JAMES H. BAXTER (introduced by DONALD D. VAN SLYKE). *Hospital of The Rockfeller Institute for Medical Research, New York.* On the basis of previous observations in rats, it was suggested that pyridine liver necrosis might be due to disturbance of circulation through the lobules, resulting in anoxia of the centro-lobular cells, rather than to direct necrotizing action (Am. J. Path., in press), and that renal cortical necrosis of choline deficiency might likewise result from circulatory disturbances (J. Nutrition, **34**, 333, 1947). Further observations, indicated below, are at least compatible with these suggestions. Necrosis occurred earliest in those portions of the functional units furthest removed from the source of blood supply, while cells about portal triads in the liver, and the glomeruli, and structures in the "lesser" renal circulation escaped. It was demonstrated by India ink injections that little circulating blood entered the involved areas. Dead cells were slowly resorbed. Necrotic regions became engorged with deoxygenated blood; involved organs enlarged, dark and tense. In terminal stages of irreversible damage, marked obstruction to perfusion of fluid through the injured organ as a whole occurred. Shock often preceded death due to liver necrosis. Observations at various stages of injury have not definitely established the immediate cause of the circulatory disturbance, or that the disturbance precedes necrosis. Decapsulation of the organs has not produced conclusive protection. Others (Himsworth, Hartroft) believe compression of vessels by swollen but viable parenchymal cells initiates necrosis under similar circumstances The circulatory disturbances, it appears, if not actually initiating necrosis, may at least be responsible for extension of the necrosis and further damage to the organism.

Correlation of the unitarian or trophoblastic thesis with the biological test of malignancy. HOWARD H. BEARD. *Department of Biochemistry, The Chicago Medical School, Chicago, Ill.* The correct etiology of malignancy and nature's remedy for it were first announced in 1902 by John Beard of Edinburgh. His trophoblastic thesis states that the wondering germ cells of early life can later be activated to divide and produce trophoblast cells which, outside the canalization of pregnancy, are the malignant cells. Since the pregnancy trophoblast begins to disappear about the time the fetal pancreas develops he also suggested the use of pancreatic extracts for the treatment of human malignancy. It is most unfortunate for the human race that these extremely important discoveries had to lie dormant until Gurchot, Krebs and Krebs recently confirmed and greatly extended the theories and treatment of malignancy advocated by Beard. The Ascheim-Zondek test is positive in most genital cancers and in some nongenital ones. Our biological test is a micro-AZ test and we have obtained positive results in 130 of 134 malignant urines tested in the last 2 years. Since the malignant (or pregnancy) trophoblast gives off syncytial steroids and chorionic gonadotrophin, these malignant hormones are extracted from the urine by alcohol-ether and, after removal of the solvents, these extracts cause hypertrophy of the spleen and/or gonads 2½ days after injection into immature rats. It is, therefore, concluded that the results of these biological tests constitute the strongest experimental evidence for the validity of the unitarian or trophoblastic thesis of malignancy.

Effect of pH on the aerobic metabolism of rabbit bone marrow. ROBERT M. BIRD and JOHN D. EVANS (introduced by WILLIAM H. SUMMERSON) *Department of Physiology, Cornell University Medical College, New York, N. Y.* The aerobic metabolism of the bone marrow cells of New Zealand White rabbits had been studied *in vitro* in

bicarbonate-containing media within the pH range 7.6 to 6.3. Between pH 7.4 and 7.1 oxygen consumption averaged 3.3 microliters per mgm. of cell protein per hour and showed little alteration with change in pH. From pH 6.8 to 6.3 oxygen consumption averaged 2.7 microliters per mgm. per hour and again was little influenced by pH changes within this range. At these two rates of oxygen consumption the average R. Q.'s were 0.97 and 0.95 respectively. Aerobic acid production was more affected by change in pH, being depressed in a substantially linear fashion with increasing acidity. At pH 7.4 it averaged 3.9 microliters per mgm. per hour; at pH 6.3, 1.4 microliters per mgm. per hour. Lactic acid formation accounted for approximately 90% of all acid produced aerobically throughout the pH range studied. Glucose utilization, in the majority of experiments, was sufficient to account for both lactic acid formation and total oxygen consumption. These findings are correlated with prior studies on bone marrow metabolism.

Effects of pH and bicarbonate on brain tissue respiration and anaerobic glycolysis. MARION K. BIRMINGHAM (by invitation) and K. A. C. ELLIOTT. *Montreal Neurological Institute, McGill University.* Previously it has been shown (Fed. Proc. **6,** 249) that there is a broad optimum pH for brain tissue respiration at about 7.4, and that apparent high optima found with slices are due to the fact that the pH of the slice is not the same as that of the medium. The anaerobic glycolysis of isotonic brain tissue suspensions has now been found to show a rather sharp optimum at about 6.8. With careful equalization of pH it was found that, compared to unbuffered Ringer solution, bicarbonate-buffered Ringer causes a slight inhibition of respiration, about 7% with 0.02 M bicarbonate-5% CO_2, about 16% with 0.04 M bicarbonate-10% CO_2. Small amounts of bicarbonate added to phosphate buffered medium in the presence of alkali papers appear to increase the oxygen uptake rate but this can be accounted for by an artefact arising from changing CO_2 tension. The rates of glycolysis in phosphate and bicarbonate buffered media of equal pH are about the same, and lactic acid accounts approximately for all of the acid produced in either medium. A large difference between bicarbonate and phosphate buffered media reported previously (Elliott and Henry, J. Biol. Chem. **163,** 361) was due to an error. Increasing bicarbonate concentration with constant CO_2 tension depresses the rate of glycolysis, but this can be accounted for by the increase in pH above the optimum.

A microanalytical method for acetic and other volatile acids. SIMON BLACK (introduced by E. S. GUZMAN BARRON). *Chemical Division, Department of Medicine, The University of Chicago.* A simple microdiffusion and microtitration procedure for volatile acids has been substituted for the usual steam distillation and macro- or semi-microtitration. Quantities of acetic acid between 10 and 100 micrograms, or equivalent amounts of other volatile acids, can be determined with an average error of less than 2 micrograms. Large numbers of analyses can be performed simultaneously. To determine acetic acid in the absence of interfering substances 1 ml. of a protein-free filtrate of the sample is made slightly alkaline and evaporated to dryness (105°) on the bottom of a 30 ml. bottle. A small glass cup (12 mm. O.D., 5 mm. deep) containing 25 c.mm. of 0.1 M $NaHCO_3$-15% NaI is centrally placed on the floor of the bottle. One-half ml. of 15 N H_2SO_4 saturated with Ag_2SO_4 is pipetted onto the floor of the bottle outside the cup. A cap with a tinfoil-covered rubber gasket is screwed on tightly and the bottle placed in an oven (105°) over night. Thirty-five c.mm. of 0.1 M KH_2PO_4-0.03% Duponol PC is added to the center cup (removed from bottle) to eliminate CO_2, then 3 drops of saturated phenolphthalein solution (in 4% EtOH) and a short (5 mm.) glass-covered iron wire (magnetic stirrer). The solution is titrated over a revolving magnet with 0.1 N NaOH from a microburette. The latter is constructed from a tuberculin syringe and a micrometer caliper. Acetic acid is calculated from the difference between sample and blank titrations.

Synthesis of fatty acids in rat liver slices. KONRAD BLOCH and W. KRAMER (by invitation). *Department of Biochemistry and Institute of Radiobiology and Biophysics, University of Chicago.* Incubation of rat liver slices in the presence of labeled acetic acid leads to extensive incorporation of isotope into cholesterol, but little isotope enters into fatty acids under these conditions (J. Biol. Chem. **162,** 441 (1945). Present experiments show that fatty acid synthesis in surviving rat liver is greatly stimulated by addition of pyruvate to the medium. Fatty acids formed from $CH_3C^{14}OONa$ contain 4–8 times more C^{14} in the presence than in the absence of pyruvate. The same stimulatory effect is shown by oxaloacetate but not by glucose, fumarate, malate or succinate. In bicarbonate buffer more than twice as much isotope is incorporated into fatty acids than in phosphate buffer. In the absence of calcium ions fatty acid synthesis is reduced to insignificant levels. Under conditions which are favorable for fatty acid synthesis, the rate of cholesterol synthesis is diminished. Utilization of acetate for cholesterol formation is reduced to less than half of the control value by addition of pyruvate or oxaloacetate.

The inhibitory effects of α-amino acids on phosphatase activity. OSCAR BODANSKY and NORMA STRACHMAN (by invitation). *Department of Pharmacology, Cornell University Medical College, New York City.* It was previously shown that glycine, in concentrations higher than the 0.006

M necessary in conjunction with other conditions for optimal activity, inhibited bone and intestinal phosphatases, the former entirely in a non-competitive manner and the latter largely so (Bodansky, O., J. Biol. Chem., **165**, 605 (1946)). The inhibition was dependent on the presence of the free carboxyl and amino groups of glycine. In the present report, the inhibitory effects of varying concentrations of other α-amino acids on phosphatase activity is studied at 25°C., at optimal alkaline pH, at optimal concentrations of Mg^{++} and of glycine (0.006 M) and at near-maximal or maximal concentrations of the substrate, Na β-glycerophosphate. The reaction velocities of rat intestinal, human intestinal, rat bone and cat bone phosphatases were reduced to 50% of their uninhibited values by, on the average, 0.11 M dl-alanine or 0.05 M glycine. Although there was some variability, there appeared to be no significant difference, for either amino acid, between the effects on the bone and intestinal phosphatases of these species. Other α-amino acids not only exerted inhibitory effects of a different order of magnitude, but also showed differences with respect to rat bone and intestinal phosphatases. The following concentrations of amino acids produced 50% inhibition in rat intestinal and bone phosphatases, respectively: 0.03 M and 0.09 M l-glutamic acid; 0.21 M and 0.01 M l-lysine; 0.006 M and 0.003 M l-histidine.

The role of bicarbonate in glutamic acid metabolism. Ernest Borek and Heinrich Waelsch. *Departments of Biochemistry, New York State Psychiatric Institute and Columbia University.* Bicarbonate has been shown to interfere with glutamic acid metabolism in *Lactobacillus Arabinosus*, apparently by blocking the amidation to glutamine (Waelsch, Prescott and Borek, J. Biol. Chem. **171**). The amount of bicarbonate needed for complete growth inhibition varies with the pH of the medium and with the glutamic acid concentration. At pH 6.3 and glutamic acid concentrations of 40, 80 and 120 gamma per ml., 1.2, 2.4 and 2.8 mgm. of bicarbonate per ml. inhibit growth completely (molar ratios 54, 54 and 40). At this pH the addition of 0.4 gamma of glutamine per ml. to the medium containing 120 gamma of glutamic acid overcomes the growth inhibition caused by 2.8 mgm. of bicarbonate. Bicarbonate also inhibits if ketoglutaric acid is substituted for glutamic acid and this inhibition is also overcome by small amounts of glutamine. 2 mg. of cysteine per ml. overcome completely the inhibition induced by 2.8 mgm. of bicarbonate per ml. The possible role of carbonic acid as the regulator of the utilization of glutamic acid either for amidation or for transamination is discussed. Observations on this mechanism in mammalian tissues are presented.

Protein and peptide turnover with respect to lysine in guinea pig liver homogenate. Henry Borsook, Clara L. Deasy (by invitation), Jacob W. Dubnoff, C. T. O. Fong (by invitation), William D. Fraser (by invitation), A. J. Haagen-Smit (by invitation), Geoffrey Keighley (by invitation), Peter H. Lowy (by invitation). *William G. Kerckhoff Laboratories of the Biological Sciences, California Institute of Technology, Pasadena 4, California.* Protein and peptide turnover in guinea pig liver homogenate was studied with the aid of lysine synthesized with C^{14} in the ϵ-position. The homogenate was incubated with an amino acid mixture (final concentration 1%) of the composition of casein in which all the added lysine was labelled, and α-ketoglutarate (final concentration 0.01 M). The pH varied from 7.5 to 9.0 in different experiments. After 6 hours at 38° under oxygen the proteins were removed and a peptide fraction containing lysine was isolated as the phosphotungstate. Its insolubility in acetone distinguishes it from the phosphotungstate of lysine. Isolation and purification procedures excluded the adsorption of unbound radioactive lysine. The lysine isolated from the protein after hydrolysis contained a small amount of radioactivity, indicating less than 0.1% turnover. The peptide fraction contained 5 per cent of the added lysine. The turnover in this fraction was therefore very high. Chromatography of the peptide fraction on filter paper gave, before hydrolysis, one spot with ninhydrin; after hydrolysis, 6 or more spots. The free α-amino nitrogen before and after acid hydrolysis indicated that there might be approximately 8 peptide bonds. The quantity of the peptide fraction available at present is too small for a complete amino acid analysis; indications are that it contains lysine, arginine, histidine, and mono-amino acids. Tryptophane and tyrosine are absent. A similar peptide fraction is normally present in guinea pig liver.

Inactivation of trypsin by acetylation and iodination. Donald E. Bowman. *Department of Biochemistry and Pharmacology, Indiana University, School of Medicine, Indianapolis, Ind.* Acetylation of trypsin by means of ketene decreases its proteolytic activity and the hypotensive effect of its intravenous administration. With increasing acetylation the decline in proteolytic activity is no less rapid than the decrease in hypotensive effect. This is in contrast with the more rapid loss of hypotensive activity which results from iodination as reported earlier. Loss of activity upon acetylation does not appear to be due to the blocking of amino groups. The tyrosine color value decreases to about half of that of untreated trypsin when the hypotensive and proteolytic activities have decreased to one tenth of their original values. Considerable recovery of proteolytic activity as the result of mild alkaline hydrolysis of acetyl trypsin

indicates that loss of this activity is at least in part associated with the linkage of acetyl groups to oxygen such as that of the phenolic hydroxyl groups. Iodination of trypsin with constant amounts of iodine less than that required to satisfy all groups has less influence on the hypotensive effect with a decrease in pH which slows the rate of iodination of groups such as histidine more than that of tyrosine. Groups which iodinate rapidly at decreased pH levels such as cysteine and tryptophane do not appear to be involved in inactivation. Failure of the two types of activity of the enzyme to remain parallel is in accord with the ability of navy bean trypsin inhibitor to inhibit proteolysis without preventing a fall in blood pressure following intravenous injection.

The glucose content of the rat carcass. MAURICE BRUGER, SAMUEL MEMBER (by invitation) and ESTELLE GOLDMAN (by invitation). *From the Medical Research Laboratory, Department of Medicine, New York Post-Graduate Medical School and Hospital.* The total glucose content of the carcass was determined in 98 young rats (Wistar strain) as control observations for studies on alloxan diabetes. Detailed precautions were taken to avoid glycolysis and the whole animal macerated in a special crusher (packed in dry ice). Somogyi filtrates were prepared and glucose determined by the Shaffer-Hartmann method. Subgroups were studied in which the pre-analytic preparations differed as follows: (a) Purina Chow Checkers or low carbohydrate diet (b) catharsis (c) evisceration (d) 24-hour, overnight or no fasting. Carcass glucose decreased as the starvation period was prolonged; fasting for 24 hours gave a high mortality rate in young rats. Purina chow was too high in carbohydrate content and gave irregular results because of variable amounts of food remaining in the gastro-intestinal tract even after catharsis (castor oil). Eviscerated animals (stomach and intestines removed) gave the highest glucose values averaging 99.0 ± 12.2 mgm. per 100 grams of rat. The most consistent results in the whole animal were obtained in a group of 34 rats ranging in weight from 39.8 to 66.8 grams on a low carbohydrate diet (J. Nutrition **10**:507, 1935, diet 10) for one week and fasted overnight. In this group, the average glucose content of the carcass was 53.0 ± 19.2 mgm. per 100 grams of rat.

Reversible oxygenation, irreversible oxidation, and decarboxylation of cobalt-amino acid complexes with oxygen gas. DEAN BURK, and (by invitation) SILVIO FIALA, JOHN HEARON, MARIE HESSELBACH, HILTON LEVY, and ARTHUR L. SCHADE. *National Cancer Institute, National Institute of Health, Bethesda, Maryland, and Overly Biochemical Research Foundation, New York.* Various cobaltous-amino acid complexes, in addition to cobaltodihistidine and certain cobaltous histidine derivatives reported on previously (J. Biol. Chem. **165**: 723 (1946); Fed. Proc. **6**: 242 (1947)), react reversibly with oxygen gas under physiological conditions. Reversibly oxygenated, yellow-brown cobaltous compounds were readily formed with lysine, arginine, tryptophane, proline, ornithine, serine, asparagine, glutamine, glutamate, glycine, and glycyl-glycine at concentrations of the order of 0.01 to 0.1 M. However, the oxygenated compounds were transformed spontaneously into pink, irreversible oxidation forms, many times more rapidly than was oxy-bis(cobaltodihistidine). In general, the stability of the oxygenated forms decreased with increasing temperature and pH. Irreversible O_2 consumption, but no intermediate reversible oxygenation, was observed with cysteine, methionine, alpha-alanine, and putrescin, and no oxygen consumption at all with beta-alanine.

The irreversible reaction is complex, and may be shown with histidine to involve several recognizable phases, including 1) an initial irreversible rearrangement of the reversible form without additional O_2 uptake, 2) eventual absorption of a second molecule of O_2 by oxy-bis(cobaltodihistidine), and 3) especially with excess amino acid, production of CO_2 and OH^-, in ratios of CO_2/O_2 and OH^-/O_2 approaching maxima of 0.5 and 1 respectively, indicating complex organic oxidation and decarboxylation. With excess cobalt it was possible to obtain complete conversion to a pink irreversible form without notable production of CO_2 or OH^-, indicating non-oxidation of the cobaltous to cobaltic state here, a result further confirmed polarographically, where no change in height or potential of the cobaltous-cobalt wave was observed upon conversion of cobaltodihistidine to oxy-bis(cobaltodihistidine) and then to the pink irreversibly oxidized complex.

A study of the microbiological assay of valine with estimation of precision of the method. MURIEL M. BURR and W. A. CRANDALL (introduced by L. I. PUGSLEY). *Food and Drugs Division, Department National Health and Welfare, Ottawa, Canada.* A synthetic medium of pure amino acids and vitamins is described for the assay of valine, employing Lactobacillus arabinosus as the test organism. Comparisons are made with other proposed methods using this organism. Comparable results are obtained in the assay of various protein hydrolysates, pharmaceuticals and proteins and recovery experiments were satisfactory. An estimation of the precision of the method is carried out according to Crandall and Burr "Precision of microbiological assays for riboflavin, niacin and pantothenic acid." (in press).

The metabolism of peptides of asparagine. C. E. CARTER (Introduced by J. P. GREENSTEIN). *Clinton National Laboratory, Oak Ridge, Tennes-*

see. Enzymes capable of hydrolyzing the peptide bond of glycyl l-asparagine without attacking the amide group have a high activity and wide spread distribution in mammalian tissues. The optimum effect exhibited by this system is at pH 8. Liver and kidney extracts are the most potent sources of the enzyme, and in dilute solution effect the complete hydrolysis of the peptide in 2.8×10^{-3} M substrate in 20 minutes. Dialysis does not inactivate the enzyme system and there is no specific metal activation, or anion effect such as has been described in the desamidation of glutamine and asparagine. Leucyl Asparagine is metabolized at the same rate as glycyl asparagine. Chloroacetyl asparagine, α-bromisocapronyl asparagine, are metabolized by aqueous tissues extracts but at a much slower rate. Hydrolysis of the peptide bond has been followed manometrically by the chloramine T reaction and by the ninhydrin reaction. A purification of the enzyme system has been effected from rat liver by ammonium sulfate fractionation of aqueous extracts.

Influence of dietary and physical factors on the urinary excretion of thiamine and pyramin. W. O. CASTER (by invitation), OLAF MICKELSEN and ANCEL KEYS. *Laboratory of Physiological Hygiene, University of Minnesota, Minneapolis.* One year 4, 4, and 2 normal young men were maintained on constant thiamine intakes of 0.6, 1.0, and 1.8 mgm. per day respectively for 5.5 months. In another year 6 and 6 men were on 1.0 and 2.0 mgm. of thiamine per day respectively for 8 months. The 24 hour urinary thiamine and pyramin excretions were followed throughout each period. On separate occasions each lasting 5 days while the thiamine and caloric intakes were kept constant, a diet providing 200 grams of protein, and one supplying 80% of the calories from carbohydrate were fed. In both of these periods, the urinary thiamine and pyramin excretions showed no consistent and significant changes over that of the preceding control period. A diet providing 75% of the calories from fat was fed for 5 days one year and for 15 days the second year. In both cases there was an indication of a significant increase in the thiamine and pyramin excretions. This significance, however, was apparent only when the factor of individual variation was eliminated from the calculations. During the second year 3 men from each thiamine intake group were subjected to a 3-day period of work at such a rate that 6000 calories were required for daily equilibrium. The other men were on the same work and dietary regimen for 10 days. Both experiments showed no significant change in thiamine excretion whereas the pyramin excretion increased significantly during the first 2 to 3 days, followed by a decline to below the control levels.

Some properties of β-keto acid carboxylases from plants. J. CEITHAML (by invitation), M. C. GOLLUB (by invitation), J. SPECK (by invitation) and B. VENNESLAND. *Department of Biochemistry, University of Chicago.* Previous observations on the occurrence of oxalacetic carboxylase in various plant preparations have been extended. The enzyme from black and red radishes, carrots and parsnip has been shown to be independent of the pyruvic carboxylase which is present in all the crude extracts from these sources. The oxalacetic carboxylase from parsley root has been subjected to a detailed study. The ability of a number of cations to activate this enzyme has been demonstrated. Mn^{++} is the best activator, but Cd^{++}, Co^{++}, Zn^{++}, Ca^{++}, Pb^{++} and Mg^{++} are also capable of showing cofactor effects. The enzyme is rapidly destroyed at room temperature on exposure to hydrogen ion activities below pH 5 or above pH 9. The active protein has been subjected to a partial purification and studies of the purified fractions with regard to their content of biotin, malic dehydrogenase and isocitric dehydrogenase are in progress and will be reported.

Repletion of protein depleted dogs with various proteins and protein hydrolysates. BACON F. CHOW and SHIRLEY DEBIASE (by invitation). *Division of Protein Chemistry, The Squibb Institute for Medical Research, New Brunswick, New Jersey.* Dogs were depleted of plasma proteins on a diet free from proteins but adequate in vitamins and calories for a period of 4 to 6 weeks. Attempts were made to replete these animals with a diet containing hydrolyzed and undigested proteins with different amino acid patterns and biological values, with a daily intake of 0.35 grams nitrogen per kgm. body weight. The total circulating plasma proteins were determined before depletion, after depletion, and during repletion. Plasma albumin decreased from 45% to 25% of the total plasma protein. The results on repletion confirm our previous findings that casein hydrolysate will regenerate both plasma albumin and globulins, whereas, lactalbumin hydrolysate will regenerate mainly albumin. Supplementation of lactalbumin hydrolysate with amino acids to give a composition similar to casein hydrolysate or supplementation of casein hydrolysate with methionine to increase the nitrogen retention to that of lactalbumin hydrolysate did not alter the plasma protein regeneration properties of the original hydrolysates. Several whole protein mixtures such as egg albumin, Melactin (a mixture of casein and lactalbumin) and wheat gluten were also fed to the depleted dogs. It was found that wheat gluten did not stimulate any appreciable regeneration of plasma proteins at this level of intake. The responses of the depleted dogs to the diet of either Melactin or egg albumin, in spite of their high biological values, is much slower than casein or lactalbumin hydrolysate.

The distribution of amino acids between fetal and maternal extracellular fluids and cells. HALVOR N. CHRISTENSEN. *The Children's Hospital, and the Department of Biological Chemistry, Harvard Medical School.* A comparison has been made of certain amino acid concentrations in fetal and maternal plasma and tissues. In the guinea pig the levels of glycine and of non-glycine, non-glutamine ("residual") α-amino nitrogen (by ninhydrin) was about five times as high in the fetal plasma (umbilical cord blood) as in the maternal plasma. The concentrations of glycine in the muscle cells of both the fetus and mother were about nine times the concentrations in their respective plasmas and the concentrations of "residual" α-amino nitrogen in the muscle cells of both organisms were about 6 times the concentrations in the respective plasmas. Thus the muscle cells of the fetus contain about 4 times as high a level of amino acids as the muscle cells of the mother, due, however, to the concentrating of amino acids by the placenta rather than to an unusual ability of fetal muscle to concentrate amino acids. The administration of glycine by mouth to the pregnant animal caused a larger increase in the glycine of the fetal plasma than of the maternal plasma. The response of the concentrating ability of the placenta to the administration of amino acids was analogous to the response of the cells of liver and muscle. These observations have a bearing on the rapid protein synthesis by the fetus.

The mechanism of enzymatic synthesis of citrulline from ornithine. PHILIP P. COHEN and SANTIAGO GRISOLIA (by invitation). *Department of Physiological Chemistry, University of Wisconsin Medical School.* Glutamic acid has previously been shown to play a specific role in the conversion of ornithine to citrulline in rat liver homogenates (Cohen and Hayano, J. Biol. Chem., **170**, 687, 1947). More recently it has been shown that washed residue of rat liver homogenate contains the enzyme system catalyzing this reaction (Cohen and Hayano, J. Biol. Chem., in press). Present studies on the mechanism of this reaction using washed rat liver residues supplemented with adenosine triphosphate, adenylic acid, magnesium and inorganic phosphate ions, indicate that glutamic acid acts as the acceptor for CO_2 and ammonia to form an intermediate compound which reacts with ornithine to form citrulline. The fixation of CO_2 and ammonia by glutamic acid requires oxygen while the transfer reaction proceeds anaerobically. Replacement of glutamic acid by glutamine, fumarate, succinate, α-ketoglutarate, aspartate, oxalacetate, isocitrate and pyruvate gave the following percentage yields of citrulline, representing glutamic acid as 100 per cent: 100; 19.2; 17.6; 3.7; 0; 0; 0; 20. Attempts to demonstrate direct fixation of CO_2 and ammonia by ornithine have been unsuccessful. The nature of the intermediate compound of glutamic acid and CO_2 and ammonia is at present under investigation.

The action of insulin on glycogen reserves. GEORGE L. COLLINS, JR. (by invitation), EDWARD M. BRIDGE, and BARBARA MAITLAND (by invitation). *From the Statler Research Laboratories of the Children's Hospital, Buffalo, and Department of Pediatrics of the University of Buffalo, Buffalo, New York.* The action of insulin in stimulating the deposition of muscle glycogen is well established. While numerous experiments have been reported showing that a similar stimulation to glycogen deposition does *not* occur in the liver, there are differences of opinion regarding the interpretation of the findings. Bouckaert and de Duve (Physiol. Rev., **27**: 39–71, January 1947) believe that the failure of insulin preparations to stimulate glycogen deposition in the liver is due to impurities in the insulin used. Others interpret the findings as a direct effect of the hormone itself. Experiments will be presented showing the effect of insulin preparations of varying degrees of purity on liver and muscle glycogen. The effect of endogenous hyperinsulinism (pancreatic adenoma) on glycogen stores will also be demonstrated.

Changes in the composition of the blood of rats fed L-histidine. H.R. CROOKSHANK (by invitation) and CLARENCE P. BERG. *Biochemistry Department, Medical College of Alabama and the Biochemical Laboratory, State University of Iowa.* The course of histidine metabolism was studied in previously fasted rats by assaying the blood for histidine, total imidazole, and amino, urea, and total non-protein nitrogen, in some instances before, and in others at various intervals after feeding single doses of L-histidine. All of these components but the urea attained maximal concentrations in 1 hour after the feeding. In 6 hours, free histidine had disappeared and total imidazole had returned to the fasting level. Amino and non-protein nitrogen diminished fairly rapidly in this period, then more slowly until the fasting levels were attained (in 12 to 16 hours). The urea rose perceptibly in the 2nd hour, became maximal in 4 to 8 hours, then fell gradually to its initial minimum in 12 to 16 hours. The small differences noted in these tests between the concentrations of histidine and total imidazole seem to preclude any considerable conversion of L-histidine to intermediates containing the imidazole ring. On the other hand, the differences between the increases in amino nitrogen found by assay and the increases attributable to histidine seem sufficient to warrant assuming either that histidine is converted into a metabolite which retains the amino nitrogen, or that, under the stimulus of the histidine, amino nitrogen is produced from some other source. The data seem

consistent with the theory that L-histidine is converted normally, at least in part, to glutamic acid. Studies in progress involve further tests on L-histidine and similar tests on D-histidine.

Dietary influence on the amino acid composition of proteins. FRANK A. CSONKA. *From the Bureau of Human Nutrition and Home Economics, Agricultural Research Administration, United States Department of Agriculture, Washington.* It was previously shown that the cystine and methionine content of a hen's egg may be increased by feeding a high protein casein diet (Csonka, Denton, and Ringel. J. Biol. Chem., **169**: 259 [1947]). Both crystalline egg albumin and conalbumin have been now prepared from individual eggs laid by hens fed different diets, and their cystine, methionine, and total nitrogen contents determined. The results show that the amino acid composition of these proteins is influenced by the hen's diet. It was also found that the nitrogen content of the crystalline albumin obtained from eggs produced by hens fed high protein diets was higher than that produced on low protein diets. The conalbumin of eggs produced by hens fed a high protein soybean diet contained a higher nitrogen content than when fed at a lower protein level. The lowest nitrogen content was observed when the eggs were produced on a high protein casein diet. The cystine and methionine percentages in both egg white proteins were not constant, and significant differences were observed depending on the dietary regimen. The cystine and methionine contents of the crystalline albumin were consistently higher in the samples prepared from eggs laid by the same hens fed a low protein diet as compared with those receiving the high protein casein diets. A significant drop in methionine percentages occurred in both egg albumin and conalbumin when the diet was changed from low to high protein soybean diet.

Critique of methods for determination of riboflavin in urine. ELMER DE RITTER (by invitation), MARY E. MOORE (by invitation), ERICH HIRSCHBERG (by invitation), and SAUL H. RUBIN. *Nutrition Laboratories, Hoffmann-La Roche, Inc., Nutley, N. J.* Riboflavin assays by several fluorimetric and microbiological procedures have been compared for human urines, ranging in content from high to vanishingly small amounts. The recent fluorimetric method of Slater and Morell (Biochem. J., **40**, 644, 1946) yielded slightly lower results for normal urines than a Florisil adsorption procedure, which, in turn, gave lower values than the double reduction method previously reported from this laboratory (Ind. Eng. Chem., Anal. Ed., **17**, 136, 1945). However, the differences between the Slater-Morell values and those obtained by the other fluorimetric methods were much smaller than those found by Morell and Slater (Biochem. J., **40**, 652, 1946) with similar methods. This better agreement was due to the fact that the present modifications of the Florisil and double reduction procedures included a permanganate treatment in both cases and the use of a *minimal* amount of hydrosulfite for blank determinations in the double reduction procedure. Microbiological assays, even those calculated by the slope-ratio method, were less reproducible than fluorimetric assays, ranging between the Slater-Morell and the Florisil values.

High levels of urea in association with normal riboflavin levels caused slight lowering of titrimetric microbiological values, but did not affect the turbidimetric measurements. High urea/riboflavin ratios in *low*-riboflavin urines, collected during total intravenous feeding of protein hydrolysates (Fed. Proc., **6**, 287, 1947), led to serious errors in titrimetric assays. For this reason, as well as for speed and convenience, turbidimetric measurement is preferred for the assay of urine.

The breakdown of free and combined adenosine-5-phosphate (AdPh) in intact human erythrocytes. ZACHARIAS DISCHE. *Department of Biochemistry, College of Physicians and Surgeons, Columbia University, New York.* In washed human erythrocytes kept for 15 minutes at 37° in presence of $\frac{M}{40}$ NaF, $\frac{M}{100}$ Na bromo acetate and glucose ATP disappears completely while glucose is phcsphorylated to hexosediphosphate and AdPh is formed. If these erythrocytes are kept for 6 days at 0° 50–80% of AdPh disappears and simultaneously hexosediphospbate and triosephosphate are formed. Their amount accounts for about 60% of disappearing ribose. When erythrocytes are treated in the same way without addition of glucose some adenylpyrophosphate remains inside cells during all the time of the experiment. AdPh disappears, though less than in presence of glucose. Hexosediphosphate + triosephosphate account now for up to 100% of disappearing pentose. No significant amounts of hexosemonophosphate were found in either tỳpe of experiment. These findings suggest, that in human erythrocytes AdPh is split in triosephosphate and a dicarbon compound. The latter is synthesized in presence of ATP to hexosediphosphate. In experiments without addition of glucose the breakdown of AdPh is accompanied by its enzymatic liberation from the coenzymefraction. This was recognized by determining free AdPh and that in coenzymes by the orcinol method according to Dische and Schwartz making readings after 3′ and 20′ heating. $\frac{20' \text{ value}}{3' \text{ value}}$ is 1.22 for AdPh and 2.25 for coenzymes. Every ribosephosphate molecule breaking down in erythrocytes seems to pass finally the stage of triosephosphate.

The effect of thyroidectomy and of thiouracil

on cytochrome *c* metabolism and liver regeneration. DAVID L. DRABKIN. *Dept. of Physiological Chemistry, Graduate School of Medicine, Univ. of Pennsylvania, Philadelphia.* The experiments to be reported have disclosed an apparently significant influence of the thyroid gland on the cytochrome *c* content of all tissues, an effect which suggests a mechanism whereby thyroxine helps regulate the oxygen consumption of tissues and the whole body.

Adult rats of average starting weight of 200 gm. were used, and were maintained on a vitamin supplemented, high (31%) protein diet. In the completely thyroidectomized animals, liver lobectomy was performed 35 days after thyroidectomy, and the experiments were terminated 14 days later. In the thiouracil studies, 50 mgm. of the drug were administered daily in a measured quantity of the diet for a period of 45 days before either sacrifice or liver lobectomy. Cytochrome *c*, water, and ribose and desoxyribose nucleic acids were determined in liver before and after regeneration, and cytochrome *c* and water content were measured also in kidney cortex, heart and skeletal muscle terminally. Thyroidectomy induced a striking reduction in total body cytochrome *c*, reflected in the concentration of the pigment in individual organs and tissues. The decrease in cytochrome *c* was about twice as great in skeletal muscle as in liver (41 against 22% reduction), with intermediate decreased values in heart and kidney cortex. In the thyroidectomized rats, liver regeneration was not strikingly interfered with, although it was of smaller magnitude than normally. The administration of thiouracil produced a reduction in the cytochrome *c* of tissues of similar magnitude as after complete thyroidectomy.

The anti-ulcer activity of aluminum dihydroxyaminoacetate. R. L. DRYER (by invitation), J. KATZ (by invitation), W. D. PAUL (by invitation), and J. I. ROUTH. *Departments of Biochemistry and Internal Medicine, College of Medicine, State University of Iowa, Iowa City.* The anti-ulcer activity of aluminum dihydroxyaminoacetate (ADA) has been investigated in rats with ligated pylorus. The animals, of both sexes, ranged in weight from 150 to 300 gm. and were maintained on a stock ration. Prior to pyloric ligation all animals were starved 72 hr. with free access to water. The ligation was performed under light ether anaesthesia; recovery from this procedure was prompt and satisfactory. The excised stomachs of rats given 2 cc. of N/100 HCl, H_2O or 0.9% NaCl by mouth immediately after the ligation consistently exhibited ulceration when examined 6 hr. later. A number of animals received 240 mgm. of ADA in the 2 cc. of HCl, H_2O or NaCl. Excision and examination of the stomach was was performed 6 hr. after the above treatment. The volume of the gastric contents, free and total acidity, pH, and peptic activity was determined in all cases. Results of these experiments show that in the absence of ADA animals uniformly suffered erosion or ulceration of a more or less severe degree. In this respect, no difference could be found in the nature or quantity of ulceration produced by either HCl, H_2O, or NaCl. In the presence of ADA the incidence of erosion and ulceration was markedly reduced. No correlation could be established between degree of ulceration and either free or total acidity or peptic activity.

Dimethylthetin as a methylator of homocysteine. JACOB W. DUBNOFF and HENRY BORSOOK. *William G. Kerckhoff Laboratories of the Biological Sciences, California Institute of Technology, Pasadena, California.* A partially purified enzyme has been isolated from the liver and kidney of the rat, guinea pig, and hog, which transfers one methyl group from dimethylthetin to homocysteine. In whole liver homogenate dimethylthetin is 6 times as effective as betaine and 60 times as effective as choline in methionine formation. The dimethylthetin transmethylase is distinguished from the choline transmethylase by its stability to dialysis and from the betaine transmethylase by its stability at pH 4.7.

Structual investigation of hydroxyaspergillic acid, an antibiotic substance produced by *aspergillus flavus*. JAMES D. DUTCHER, O. WINTERSTEINER and A. E. O. MENZEL (by invitation). *Division of Organic Chemistry, Squibb Institute for Medical Research, New Brunswick, N. J.* Under certain conditions of growth, *Aspergillus flavus* produces, not aspergillic acid, $C_{12}H_{20}N_2O_2$, but hydroxyaspergillic acid, $C_{12}H_{20}N_2O_3$, which is much less antibacterial. Subsequent to the establishment of the structure of aspergillic acid as 1-hydroxy-3,6-di-*sec*-butyl pyrazone-2 [J. Biol. Chem., **171**, 321, 339 (1947)] we have investigated the structure of hydroxyaspergillic acid. Both compounds are hydroxamic acids as shown by the ferric chloride color test and formation of a characteristic copper salt. Hydroxyaspergillic acid can also be reduced by hydrazine to the neutral desoxyhydroxyaspergillic acid. Evidence that the carbon and nitrogen skeleton is the same in both compounds was obtained by sodium and alcohol reduction to comparable piperazine derivatives. The indifference of hydroxyaspergillic acid towards diazonium coupling and bromination suggested, at first, that the additional oxygen atom occupied position 5 of the pyrazine ring, but subsequent reactions failed to verify this hypothesis. Hydriodic acid reduction caused the elimination of the elements of H_2O with the formation of a product containing a non-cyclic double bond; and heating with strong phosphoric acid similarly led to a dehydro-product. The most

likely structures therefore were those in which the additional hydroxyl group occupied the tertiary position of one or the other of the secondary butyl side chains. Confirmation of the nature of the dehydro-compounds has been achieved by reduction to products identical with reduction products of aspergillic acid, but the precise location of the hydroxyl group has not been established yet. However, the formulation which places it in the side chain situated at position 6 is favored.

Measurement and study of immune precipitates by means of ultraviolet absorption spectroscopy. HERMAN N. EISEN and ALBERT S. KESTON (introduced by ROBERT C. WARNER). *Departments of Medicine and Chemistry, New York University College of Medicine.* Ultraviolet absorption spectroscopy of specific precipitates furnishes an accurate, sensitive method for the quantitative measurement of serum antibody levels. The capsular polysaccharide of pneumococcus type V and rabbit pneumococcus type V antiserum gave a precipitate whose absorption spectrum was that of the antibody protein. This spectrum was found not to change over a broad range of the precipitin curve. However, when a protein antigen (bovine serum albumin) was used with homologous rabbit antiserum, the immune precipitates showed changes in the absorption spectra in different regions of the precipitin curve consistent with the expected variations in the antigen-antibody composition of the precipitates. Analysis of spectral data offers, within limits, an opportunity to determine the composition of those immune precipitates whose components have significant and different absorption spectra.

Immunochemical studies with proteins labelled with trace amounts of radioactive iodine. HERMAN N. EISEN and ALBERT S. KESTON (introduced by ROBERT C. WARNER). *Departments of Medicine and Chemistry, New York University College of Medicine.* Proteins were labelled with I^{131} by direct iodination. The amount of iodine involved was extremely small yet highly radioactive preparations were obtained. In these preparations only an occasional protein molecule was tagged with radioactive iodine. The label was stable, and antisera against the native protein seemed unable to distinguish between labeled and unlabelled protein. Labelling of protein was also carried out by coupling with p-I^{131} phenyldiazonium chloride. The preparations so obtained also retained specificity. However, part of the label dissociated from the protein over a period of weeks. Both types of labelled protein proved capable of being used as sensitive indicators in the determination of serum precipitins.

Metabolism of cerebral cortex from different areas and various animals and from epileptic patients. K. A. C. ELLIOTT. *Montreal Neurological Institute, McGill University.* The O_2 uptake rate of slices of cerebral cortex from various animals ranging in size from mouse to man is roughly inversely proportional to the tenth root of the common body weight of the individual of the species. Though the rate varies with the composition of the suspending medium, the same relation holds in different media. Within a species, the rate does not vary greatly or consistently with the size of the individual. Slices from different areas of the cortex respire at appreciably different rates; particularly dorsal areas respire more actively than lateral in all animals studied. Usually the second slice respires somewhat more rapidly than the surface slice. Active aerobic glycolysis, usually falling off fairly rapidly, occurs in brain slices from all species if the metabolism is studied immediately after introduction of the slices into ringer-bicarbonate medium. With isotonic suspensions aerobic glycolysis is not appreciable.

The relative rates of oxygen uptake, carbon dioxide evolution, aerobic glycolysis, and anaerobic glycolysis in the presence of the optimal amount of pyruvate, were the same in isotonic suspensions and slices from all the normal animals studied. Focal epileptogenic human brain tissue, excised for therapeutic purposes by Dr. Wilder Penfield, respired, on the average, at the rate expected from the results on normal animals. Relatively high anaerobic glycolysis occurred with suspensions of all the human samples but it is not certain whether this is related to the epileptic process.

The quantitative estimation of steroid alcohols. LEWIS L. ENGEL, HELEN R. PATTERSON and HILDEGARD WILSON (introduced by A. BAIRD HASTINGS). *Medical Laboratories of the Collis P. Huntington Memorial Hospital of Harvard University, at the Massachusetts General Hospital, Boston, Massachusetts and the Department of Biological Chemistry, Harvard Medical School, Boston, Massachusetts.* Studies of the intermediary metabolism of the steroid hormones have been hampered by the absence of a simple and relatively specific method for the estimation of steroid alcohols in urinary extracts. In an effort to devise such a method it was found that hemi-3,5-dinitrophthalates of steroid alcohols develop a color suitable for analytical purposes upon treatment with methanolic potassium hydroxide. Pure steroid alcohols were treated with excess 3,5-dinitrophthalic anhydride in pyridine solution. After dilution of the reaction mixture with hydrochloric acid the half-esters which were quantitatively formed were extracted with ether, washed and transferred to methanol. For analysis, aliquots were diluted with methanol and 5 N methanolic potassium hydroxide so that the final potassium hydroxide concentration was 1.5 N. The color developed showed maximal absorption at 510 mμ

and adherence to Beer's Law in the range 0.3–1.5 μ M of ester, provided readings were made 5–15 minutes after mixing the reagents. At higher alkali concentrations sensitivity was increased but more rapid fading was encountered. No significant color was developed by 3,5-dinitrophthalic acid in 1.5 N methanolic potassium hydroxide. The absorption spectra, extinction coefficients and variation of extinction coefficients with alkali concentration were similar for all of the compounds thus far studied. Preliminary results indicate that this method may be applied to the estimation of alcohols in urinary extracts.

Influence of polyhydric alcohols on the clotting of fibrinogen. JOHN D. FERRY and SIDNEY SHULMAN (by invitation). *Department of Chemistry, Univ. of Wisconsin.* When a fibrin clot is formed from solutions of bovine fibrinogen and thrombin, its properties and structure depend markedly (as in the case of human fibrin) on the pH, ionic strength, and other conditions. At pH 6.3 in 0.45 M NaCl, a "coarse" clot (opaque, readily synergizing) is formed. The coarseness of structure, as measured by opacity, is profoundly affected by small quantities (1 to 5%) of hydroxyl compounds. Monohydric alcohols (ethyl, butyl) increase opacity slightly; compounds with 2 to 6 hydroxyls decrease opacity; compounds with many hydroxyls (polyvinyl alcohol, starch) increase opacity. The concentrations in moles OH per liter required to decrease clot opacity to a given value provide a measure of comparative effectiveness: glycerol and mannitol, 1.6; 2-methyl, 2,4-pentanediol and pentaerythritol, 0.5; tetramethylene glycol, 0.25; hexamethylene glycol, 0.2; pentamethylene glycol, 0.1. All of these substances prolong the clotting time. They do not affect thrombin activity. They have no irreversible effect on the fibrinogen; when introduced and then removed by dialysis before clotting, they cause no alteration of clot structure.

These results indicate an interaction between fibrinogen and polyhydric alcohols which depends critically on the spacing between hydroxyl groups. The interaction increases in the order $0 < 1 < 2 < 4 < 3$, where the numbers denote the number of carbon atoms between carbons bearing hydroxyls.

The quinoid stage of tyrosine metabolism. ELLA H. FISHBERG. *Biochemical Laboratory of Beth Israel Hospital, New York City.* It has recently been shown that benzoquinone acetic acid, a substance capable of *in vitro* and *in vivo* methemoglobin formation in subjects showing evidence of vitamin-C depletion. It is an obligate intermediate in the normal catabolism of tyrosine and its existence in the blood can be demonstrated by a characteristic absorption spectrum in the ultraviolet. In certain subjects where the tyrosine metabolism is interrupted at the quinoid stage, the amount of benzoquinone acetic acid excreted in the urine can be determined quantitatively by titrating with thiosulfate the amount of I released by the quinone from an acidified solution of KI. Using this value in conjunction with the density of the u.v. absorption spectrum of this urine it becomes possible by appropriate dilution to determine the concentration of benzoquinone acetic acid in the blood. The concentration of BQA in the blood is constant in the normal organism. We have encountered marked deviations only in those conditions in which some abnormality of liver function may be clinically demonstrated or experimentally induced.

Blood plasma anti-glucuronidase activity. WILLIAM H. FISHMAN, KURT I. ALTMAN (by invitation) and BELLE SPRINGER (by invitation). *Departments of Surgery and Biochemistry, University of Chicago, Chicago, Ill.* β-Glucuronidase activity is strongly inhibited in the presence of untreated or boiled mammalian blood plasma or serum. Only a slight decline in inhibitory activity is observed with plasma dilutions of 1 to 5 or 10. With greater plasma dilutions the inhibitory activity diminishes more markedly, although demonstrable effects are still evident at 1 to 100 dilution. The activity of several other enzymes is not affected by boiled plasma suggesting a measure of specificity to the glucuronidase inhibiting principle. Body tissues contain very much less glucuronidase inhibitor activity than blood. Very frequently also, glucuronidase activity is inhibited in the presence of urine although here much of the inhibition disappears after dialysis. In plasma, glucuronidase inhibiting power is only slightly affected by dialysis either before or after boiling. However, when acidified plasma is boiled, the inhibiting activity is removed with the precipitated protein. From these observations it would appear that plasma glucuronidase inhibition may be a property of a large, heat-stable, non-dialyzable molecule. It is proposed that the glucuronidase inhibition of blood plasma may be due to an anti-enzyme which possibly may play a role in the regulation of tissue glucuronidase activity.

Alkaline phosphatase activity of intestinal lymph of the rat. EUNICE V. FLOCK and JESSE L. BOLLMAN. *Mayo Foundation, Rochester, Minnesota.* The intestinal mucosa has been considered as a possible source of the alkaline phosphatase of plasma largely owing to its high content of phosphatase and the observation of a decrease in plasma phosphatase following fasting and an increase after certain meals. Analysis of the intestinal lymph provides more direct evidence for this function of the intestinal mucosa.

The intestinal lymphatics of rats were cannulated with polyvinyl plastic tubing. A control sample of lymph was collected overnight, a meal given by stomach tube, and the lymph collected

continuously. Analysis of the lymph collected was made at the 2nd, 4th, 6th, 12th, and 24th hours. Following a fat-free meal the alkaline phosphatase activity of the lymph increased 3 to 6 times the control values by the 6th hour and remained high through the 12th hour after which the activity returned to the original value. Following a similar meal to which fat had been added the phosphatase activity was similar to that obtained with the fat-free meal alone. The phosphatase activity of plasma collected from fed rats after 2 days of lymph drainage was as low as that in normal rats fasted for this period of time.

Brain proteins: isolation of a birefringent liponucleoprotein. JORDI FOLCH and L. L. UZMAN (by invitation). *McLean Hospital, Waverley, Mass., and Harvard Medical School.* Brain tissue is treated with a 4:1 mixture of ether:CH_3OH and then with ether at —15°. The residue is extracted with water which is changed every few hours. Extracts are cleared by 12,000 rpm. centrifugation, brought to pH 8, then to 4.5, precipitate (A) that forms collected, and the filtrate $\frac{2}{3}$ saturated with $(NH_4)_2SO_4$. Any precipitate that forms is dialyzed and lyophilized (B). Material B is obtained only during the first 12 hours of extraction while A is obtained even after 48 hours. B is a protein, free of P and lipids. A is a liponucleoprotein. It is insoluble at pH 4.5 and soluble at higher and lower pH. It contains 35 per cent lipids and 1.1 per cent P. Lipids are firmly bound (prolonged boiling with $CH_3OH:CHCl_3$ being required for their removal) and are found to be a mixture of phosphatides and carbohydrate-containing lipids. The lipid-free material contains 0.72 per cent P (it accounts for half of the brain protein P) and purines with the $\frac{\text{purine}}{P}$ ratio of a nucleoprotein. Ultraviolet absorption spectrum of a solution of the isolated purines is almost identical with that of an equimolar solution of guanine and adenine of same concentration. The liponucleoportein is birefringent and its X-ray pattern (Dr. Bear, M.I.T.) shows a degree of order consistent with crystalline structure.

Fractionation of serum cholesterol. J. C. FORBES and WILLIAM B. PORTER (by invitation). *Depts. of Biochemistry and Medicine, Medical College of Virginia, Richmond, Virginia.* Experimental data will be presented showing that when normal blood serum, dried in vacuo from a frozen state, is extracted with chloroform at about 5°C. a small but constant percentage of the total cholesterol is extracted. This fraction in the case of 20 normal individuals varied from around 7 to 18%, the majority being between 10 and 13%. On the other hand the extractable cholesterol of the serum of patients with the nephrotic syndrome with marked hypercholesterolemia was very high, being in some cases well over 90%. Data will also be presented on other clinical conditions as well as on rabbits rendered hypercholesterolemic by means of a high cholesterol diet.

Concerning the mechanism of interaction of egg white trypsin inhibitor and trypsin. HEINZ FRAENKEL-CONRAT, R. S. BEAN (by invitation), and HANS LINEWEAVER. *Western Regional Research Laboratory, Albany, Calif. Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Department of Agriculture.* The trypsin inhibitor of egg white was recently identified as native ovomucoid (Hans Lineweaver and C. W. Murray, J. Biol. Chem., in press). Unpublished results from this Laboratory show that, in contrast to other trypsin inhibitors, ovomucoid has weak to no action on other proteolytic enzymes. Because of this specificity, the mode of interaction of trypsin and ovomucoid appears of particular interest. The chemistry of this interaction has been studied through the preparation of derivatives of both proteins.

The main results were: (a) Acetylation of most of the amino groups inactivated neither the inhibitor nor trypsin. But acetylated trypsin was no longer susceptible to inhibition by ovomucoid or its acetyl derivative. (b) In contrast to amino groups, most carboxyl, phenolic, guanidyl, and hydroxyl groups in ovomucoid appeared essential for its inhibiting action. (c) The proteolytic activity of trypsin was independent of its most reactive carboxyl and phenolic groups, but depended upon the integrity of amide, guanidyl, hydroxyl, and indole groups. Thus it appears that *trypsin* combines with the *substrate* through groups other than amino, but combines through its amino groups with the acid groups of the *inhibitor*. Other groups in both proteins probably perform important secondary functions in stabilizing the primary ionic combination. Limited kinetic studies indicate that inhibition of trypsin by ovomucoid is of the noncompetitive type, in agreement with the chemical indications that the substrate and inhibitor combine with different active groups of trypsin.

Oxidation-coupled phosphate exchanges in the acid-insoluble esters of cell-free liver preparations. MORRIS E. FRIEDKIN (by invitation) and ALBERT L. LEHNINGER. *Departments of Biochemistry and Surgery, University of Chicago, Chicago.* Esterification of inorganic phosphate coupled to the oxidation of malate takes place in properly supplemented suspensions of particulate material from rat liver, as demonstrated by rapid incorporation of inorganic phosphate (labelled with P^{32}) into the acid-soluble esterified fraction. In order to determine whether the oxidation-coupled esterification of phosphate in the acid-soluble fraction is functionally related to phosphate exchanges in the acid-insoluble esters, we have studied P^{32} distribution in the latter following

aerobic incubation of the suspensions in the presence of 1-malate, ATP, Mg^{++}, KCl, inorganic phosphate labelled with P_{32}, and glycylglycine buffer. After extraction of acid-soluble phosphate the acid-insoluble P was separated into phospholipid, nucleic acid and an unidentified P-containing residue ("phosphoprotein") by the method of Schneider (J. Biol. Chem., 161, 293 (1945)). The nucleic acid fraction was further separated into ribo- and desoxyribo-nucleic acid fractions. The specific radioactivity of each fraction was determined. The acid-insoluble P showed a much lower rate of turnover than the acid-soluble P, comparable to that observed in the intact liver. All three acid-insoluble P fractions showed incorporation of P^{32}. Desoxyribonucleic acid showed no significant radioactivity although the two nucleic acids are present in roughly equal amounts in the suspension. The high specific activity of the "phosphoprotein" fraction suggests an active metabolic role for the unidentified esters present.

Conversion of protein to glucose in vitamin deficiencies. OLIVER H. GAEBLER and PAUL BARTLETT (by invitation). *Henry Ford Hospital, Detroit.* The relationship between glucosuria and vitamin deficiency in experimental pancreatic diabetes may be complicated by the fact that interference with formation of glycogen or fat, or with oxidation, would increase glucosuria, while slower absorption or diminished glucogenesis from protein would decrease it (Endocrinology **39**: 239, 1946). To study glucogenesis from protein separately, D:N ratios were determined in dogs during various deficiencies. The animals received a purified, carbohydrate-free diet high in vitamin-free casein, and were phlorhizinized after depletion of one vitamin. D:N ratios were determined for 4 days during the deficiency, and for several days after supplying the missing vitamin. Ratios before and after deficiency could thus be compared in the same animal. Two dogs depleted of thiamine for 25 and 27 days respectively showed average ratios of 3.26 and 3.27; after terminating the deficiency the average ratios were 3.34 and 3.54. In dogs depleted of riboflavin for 51 and 55 days, the average ratios were 3.26 and 2.95; after administering this vitamin the average ratio in the first animal was 3.13. Results in pyridoxine deficiency have been reported in another connection (Fed. Proc. 6:236, 1947). In all experiments the characteristic effects of deficiency were present and responded dramatically to administration of the missing vitamin. None of the ratios observed during deficiencies were strikingly different from values obtained with casein by other observers, and the increases in the ratios after terminating deficiencies of thiamine and pyridoxine were not decisive.

Studies on glycolysis of mouse lymphosarcoma. HELENA GILDER (introduced by VINCENT DU VIGNEAUD), MARION H. WILSON (by invitation), JOHANNA M. LEE (by invitation). *Department of Biochemistry, Cornell University Medical College, New York, N. Y.* Aerobic and anaerobic glycolysis of the Gardner lymphosarcoma cells have been studied in the Warburg manometer. As is characteristic of tumor tissue the lymphosarcoma cells have a high aerobic and anaerobic glycolysis when optimal glucose and bicarbonate concentrations are present. Comparative studies of the effectiveness of other substrates in lieu of glucose suggest that the metabolic pathway for lactic acid production in the cells studied may not be identical with the Emden-Meyerhof-Cori metabolic pathway demonstrated in muscle and in yeast. These studies indicate substantially decreased glycolysis when certain phosphorylated carbohydrates are substituted for glucose.

Fractionation of lymphoid tissues. ERLAND C. GJESSING (introduced by ALFRED CHANUTIN). *Biochemical Laboratory, University of Virginia.* Calf thymus and mouse thymomas were fractionated by utilizing the low temperature-ethanol procedures intorduced by Cohn and associates. The stroma, lymphocytes and the saline soluble components were first separated and each was subjected to fractionation. The nucleic acid distribution for the various fractions was determined by spectrophotometric and chemical analyses. A nucleo-protein-rich fraction was obtained from the stroma. Three fractions were obtained from lymphocyte nuclei; two distinctly different nucleoproteins and a nucleic acid-free protein were precipitated. The saline soluble components were separated into 4 fractions. They were characterized as follows: The largest fraction obtained by isoelectric precipitation contained nucleic acid; two fractions were free of nucleic acid and the remaining precipitate was rich in lipides. Using the same fractionating procedures of thymus and thymomas, a number of differences in the types of precipitates were noted. These phenomona will be discussed.

Rate of $C^{14}O_2$ excretion following intraperitoneal administration of isotopic bicarbonate and acetate. R. GORDON GOULD, (by invitation, I. M. ROSENBERG, (by invitation), MAROTT SINEX, (by invitation), and A. BAIRD HASTINGS. *Dept. of Biological Chemistry, Harvard Medical School, Boston, Mass.* Fasted rats were injected with C^{14}-labeled bicarbonate in amounts of less than one milligram and the respiratory excretion of $C^{14}O_2$ followed for 4–6 hours. The results permit calculation of the specific activity of respiratory CO_2 (and therefore of blood bicarbonate) and its variation with time, as well as the extent of excretion of the C^{14}. The specific activity of respiratory CO_2 reached a maximum value within 10 minutes after injection and then decreased exponentially for approximately an hour, at a rate corresponding

to a decrease in value of 50% in 16 minutes. At about 80–100 minutes the rate of decrease changed to a considerably slower one. Carboxyl-labeled acetate of high specific activity was administered in small dosage in the same manner and half of the C^{14} was recovered as respiratory CO_2 within 40–50 minutes. The specific activity attained its maximum value in 15–25 minutes; it then decreased exponentially during the subsequent 2 hours, but at a slower rate than the initial rate observed following isotopic bicarbonate administration. Carboxyl-labeled succinate was also studied and the results, which were in contrast to those obtained with acetate, will be presented. The extent of excretion during the experimental period was over 90% for bicarbonate and somewhat less for acetate.

Xanthurenic acid excretion in pyridoxine deficient rhesus monkeys. LOUIS D. GREENBERG and JAMES F. RINEHART (by invitation). *Divisions of Pathology and Pharmacology, University of California Medical School, San Francisco.* Using the method of Miller and Baumann (J. Biol. Chem. **157,** 551, 1946) the quantitative excretion of xanthurenic acid was followed in 7 pyridoxine deficient rhesus monkeys on the diet and supplements previously described (Fed. Proc. **5:** 222, 1946). Estimations of the xanthurenic acid eliminated in urine were made before and after the administration of 0.5–1.0 gram doses of Dl- or L-tryptophane per os. Four animals were studied in this manner before and after withdrawal of vitamin B_6 from the diet, so that they could serve as their own controls. On a good intake of the vitamin only small quantities (apparent xanthurenic acid 1.5–3.0 mg.) of substances reacting with the $FeCl_3$ reagent were excreted before or after the administration of tryptophane. These values probably represent compounds other than xanthurenic acid, since this reagent reacts with phenylpyruvic acid and most likely with other alphaketo acids. As early as 18 days after withholding pyridoxine, there occurred marked increases in the xanthurenic acid excretion following the feeding of tryptophane. However, only a small portion of the total test dose of tryptophane was excreted as the chromogenic compound; for example, the average excretion expressed as per cent of the test dose is 2.1 with 0.5 gram Dl-, 3.0 with 1.0 gram DL-, and 3.8 with 0.5 gram L-tryptophane. The urine specimens of moderately or severely deficient monkeys were usually colored green and the intensity of the color was increased by the addition of ferric chloride.

Partial purification of cathepsin II from swine kidney. HELMUT R. GUTMANN (by invitation) and JOSEPH S. FRUTON. *Dept. of Physiological Chemistry, Yale University, New Haven.* Aqueous extracts of swine kidney contain a wide variety of proteolytic enzymes. In addition to the endopeptidase (cathepsin II) which hydrolyzes benzoyl-L-argininamide, there are present several exo and endopeptidases. In the crude extract, the exopeptidase activity toward substrates such as glycylglycylglycine, L-leucinamide, or carbobenzoxyglycyl-L-phenylalanine exceeds the activity of Cathepsin II. A purification procedure has been devised for Cathepsin II which permits the elimination of the known exopeptidases and the other endopeptidases for which synthetic substrates are available. This procedure involves fractional precipitation with ammonium sulfate, dialysis against distilled water, precipitation with yeast nucleic acid, selective extraction of the protein-nucleate, and finally, fractional precipitation with ammonium sulfate. The purified enzyme hydrolyzes, in addition to benzoyl-L-argininamide, gelatin, denatured hemoglobin, and salmine. As in the case of the synthetic substrate, the digestion of the proteins is favored by the presence of cysteine. When purified cathepsin II is partially inactivated by heat, the rate of cleavage of benzoyl-L-argininamide, gelatin, and salmine is decreased to about the same extent. This result supports the view that the endopeptidase activity of Cathepsin II is associated with a protein-splitting enzyme.

Further observations concerning antithiamine activity in plants. J. R. HAAG and P. H. WESWIG (by invitation). *Oregon Agricultural Experiment Station, Corvallis, Oregon.* Our previously announced studies (*Journal of Biological Chemistry,* 165, 737 and *Federation Proceedings,* 6, 408) dealing with thiamine deficiency induced in rats fed rations containing air-dried bracken fern have been continued and extended to related plant species. Bracken fern rations, compounded from a single lot of fern showed decreasing toxicities in the order of fresh fern, air-dried fern fed in moist rations, and air- and sun-dried fern. We have been unable to demonstrate any harmful effects to rats of rations containing as much as 40% of their dry matter as fresh fern steamed for 30 minutes. Although some evidence of antithiamine activity in related plants has been obtained, none so far tested have shown activity to equal that of bracken fern. Chemical and microbiological assay procedures have raised numerous questions concerning their interpretation in terms of animal performance. The Melnick and Field procedure, as applied to NaCl extracts, seems to respond to a quick-acting factor which possesses considerable heat stability. The thiochrome and microbiological procedure have so far offered more promise of results directly applicable to animals.

An infrared and X-ray study of polymorphism in 5-pregnen-3(β)OL-20-one. WILLIAM J. HAINES, MARI P. GOODWIN, GEORGE PISH and FOIL A. MILLER (introduced by MARVIN H. KUIZENGA). *Research Laboratories, The Upjohn Company,*

Kalamazoo, Michigan, and *Division of Physical Chemistry, University of Illinois, Urbana.* A preliminary report from this laboratory (paper presented at Meeting of American Chemical Society, New York City, 1947) has described the isolation of 5-pregnen-3(β)ol-20-one (pregnenolone) from hog testes. Although this natural product was readily identified by its melting point, optical rotation and the properties of its oxime-derivative, unexpected differences were observed when its X-ray powder pattern and infrared absorption spectrum (Nujol mull) were compared with those obtained for a sample of synthetic pregnenolone. However, the infrared spectra of chloroform solutions of the two samples were identical. This behavior suggested the existence of polymorphic crystals. The present study was undertaken as an attempt to produce a sample of synthetic pregnenolone which had a crystal form identical with that of the natural product. Since the latter had been prepared by chromatography over magnesium silicate-"Celite" mixtures followed by crystallization from absolute methanol, these techniques were applied to a sample of synthetic pregnenolone having X-ray pattern A and infrared *mull* spectrum I. The first chromatogram caused a conversion to an intermediate form, X-ray pattern B and infrared *mull* spectrum II. Rechromatography of this material resulted in a form having X-ray pattern C and infrared *mull* spectrum III. This fraction was spectroscopically identical with the natural pregnenolone. The melting points, optical rotations and infrared spectra *in solution* indicated the chemical identity of these three crystal modifications.

The interrelationship of these crystal forms and their mode of interconversion are considered. The role of polymorphism in the identification of crystalline compounds is discussed.

The metabolism of parenterally administered amino acids. I. glycine. PHILIP HANDLER, HENRY KAMIN (by invitation) and JEROME S. HARRIS (by invitation). *Departments of Biochemistry and Pediatrics, Duke University School of Medicine, Durham, N. C.* Glycine was administrred intravenously to female dogs, under dial anesthesia, at rates from 1 to 8 mg. N/kilo/min. Urine was collected constantly by catheter and blood samples obtained periodically. All dogs died, in respiratory and renal failure, after 600–900 mg. N/kilo had been administered; their survival time was roughly inversely porportional to the rate of administration. Samples of various organs were then removed, and frozen in acetone-CO_2. Other dogs, which received 0.4% NaCl or 1.5% Na_2SO_4 solutions for equal periods, served as controls.

The "rate of utilization" was defined as α-amino N administered — [α-amino N excreted + α-amino N in extracellular fluid] /kilo/min. and included cellular accumulation as well as oxidation, etc. The maximum rate of glycine "utilization" was found to be 2 mg. N/kilo/min. while the maximum rate of urea formation under these conditions was 1.2 mg. N/kilo/min. Accumulation of α-amino N was most marked in liver and progressively less in kidney, heart and skeletal muscle. This was entirely glycine accumulation since, in liver, there simultaneously occurred a drop of 50–75% in the concentration of other amino acids measured by microbiological assay in tungstate filtrates. The blood concentration of these amino acids remained relatively constant, but their urinary excretion increased 5–75 fold when the blood glycine concentration was markedly elevated. However, this excretion accounted for no more than 20% of the amino acids which had disappeared from liver. When glycine was administered at 1 mg. N/kilo/min., for 18 hours, urea N formation exceeded glycine N administration by about 20%. This phenomenon and the disappearance of liver amino acids are thought to be related to the specific dynamic action of glycine.

Conditions necessary for complete decarboxylation of pure L-lysine and L-tyrosine by amino acid decarboxylases. MARTIN E. HANKE. *Department of Biochemistry, University of Chicago.* Precise study in the Van Slyke-Neill manometric apparatus of the yield of CO_2 obtained from pure L-lysine or L-tyrosine with decarboxylases prepared from *Bacterium cadaveris* or *Streptococcus fecalis* respectively, has shown only 97 to 98% of theoretical. This yield of CO_2 is increased by removal of O_2 from the reaction mixture, or more conveniently and effectively, by the addition of a large excess of L-leucine or DL-alanine. Glycine or cysteine alone is entirely ineffective although cysteine does augment the effect of leucine or alanine. With 1000 micromols of DL-alanine plus 125 micromols of cysteine in 6.5 ml. of reaction mixture, the yield of CO_2 from 5 to 20 micromols of L-lysine or L-tyrosine is 99.5%.

Our present interpretation is that the decarboxylase preparations (crude extracts of bacterial cells) contain some L-amino acid oxidase, which oxidizes the L-lysine or L-tyrosine to a slight extent (2 to 3%) before decarboxylation is complete. The leucine and alanine may act by preferential reaction with the oxidase and consequent removal of O_2, or possibly by competitive prevention of access of the lysine and tyrosine to the oxidase. Added leucine or alanine with or without cysteine has no effect on the decarboxylase-CO_2 values of protein hydrolysates, since here there is an excess of other amino acids to tie up the oxidase. Decarboxylase-CO_2 values for lysine and tyrosine on hydrolysates of crystalline bovine blood albumin agree to within 1 part in 200 with those reported by the isotope dilution method.

The effect of protein quality of previous intake on the consequences of acute starvation. ROBERT A. HARTE (by invitation), JOHN J. TRAVERS (by invitation) and PETER SARICH (by invitation) (by JAMES B. ALLISON). *Research Laboratories, The Arlington Chemical Company, Yonkers, 1, N. Y.* In animal feeding experiments the common impression is that body weights are intrinsically more accurate than food intake figures. A review of physiological considerations quickly belies this impression. An exploratory experiment revealed that adult rats on a stock diet lost no appreciable amount of weight during the first 2 hr. following the removal of food. (In all experiments there was free access to water.) Thereafter weight loss was precipitous during the next 22 hr. The course of weight change during subsequent refeeding and food deprivation periods was followed. These observations prompted examination of the role of protein quality during the period before food withdrawal. A group of 135 rats whose growth had been followed for 5 wk. after weaning on synthetic isocaloric rations containing 10% (N x 6.25) of 7 different proteins were available for this study. They were weighed at intervals during 24 hr. following removal of food. Since they had attained differing weights during the growth period the data were analyzed by covariance technics to make approximate correction for this fact. In this way the effects of quality of different proteins previously ingested on the ability of the rat to withstand short time acute starvation could be assessed. When so analyzed the differences between proteins were highly significant ($p<0.001$). The proteins could be divided into 2 groups; 1 showing an average adjusted weight loss of about 7% and including whole egg, beef, egg white, and 2 casein samples; the other showing an average adjusted weight loss of about 10% and including wheat gluten and peanut flour.

Action of peptidases on β-alanine peptides. H. THEO HANSON (by invitation) and EMIL L. SMITH. *Laboratory for the Study of Hereditary and Metabolic Disorders and the Depts. of Biochemistry and Medicine, University of Utah School of Medicine, Salt Lake City, Utah.* Some years ago, Abderhalden and co-workers reported that peptides containing β-amino acids were not hydrolyzed by extracts of pancreas or intestine. We have found that certain peptidases will attack β-alanine peptides. Prolidase of rabbit muscle hydrolyzes glycyl-L-proline (GP) about 200 times as fast as β-alanyl-L-proline (BP). Moreover, the hydrolysis of GP is inhibited about 50% in the presence of an equimolar concentration of BP, indicating that both compounds combine with the active groups of the same enzyme. Leucinaminopeptidase hydrolyzes L-leucyl-β-alanine about one-fourth as rapidly as it does L-leucylglycine; the rates are not additive in the presence of both substrates, and the hydrolysis of both is activated by Mn^{++}. The enzyme which attacks glycylglycylglycine and other tripeptides has been found to hydrolyze glycyl-β-alanylglycine quite rapidly, glycylglycyl-β-alanine somewhat more slowly and β-alanylglycylglycine extremely slowly. The hydrolysis of glycyl-L-leucine is about 200 times as fast as that of β-alanyl-L-leucine. It is evident from our results with these and other β-alanine-containing substrates that the specificity of peptidases towards α-amino acids is not absolute. The hydrolysis of compounds of unknown structure by proteolytic enzymes can no longer be regarded as proof that the compound contains only α-amino acids.

Enzymatic cleavage of organic halides. LEON A. HEPPEL and VIRGINIA T. PORTERFIELD (introduced by B. L. HORECKER). *Laboratory of Physical Biology, National Institute of Health, Bethesda, Maryland.* It is known that inorganic bromide accumulates in the tissues of animals given various aliphatic bromine containing compounds. No *in vitro* studies are recorded. In the present investigation it was found that protein fractions of the livers of several animal species catalyzed the dehalogenation of a series of aliphatic halides. These included mono-bromo-mono-chloromethane, 1,2-dibromoethane, ethyl bromide, ethylidene bromide, β-bromopropionic acid, 1-chloro, 2-bromoethane and 1,2-dichloroethane. The enzymatic activity was measured either by the determination of inorganic halide or by following the rate of CO_2 evolution in a bicarbonate buffer due to the formation of fixed acid. Heating for 5 minutes at 60°C. or introducing 10^{-3}M. Hg^{++} caused complete inhibition. A five-fold purification of the crude tissue extract was obtained by fractionation with ammonium sulfate. Dehalogenation of mono-bromo-mono-chloromethane resulted in the formation of formaldehyde which was determined quantitatively by the chromotropic acid method. In addition, the crystalline dimedone derivative of formaldehyde was isolated and its melting point determined.

The rates of absorption of l- and dl-methionine. W. C. HESS. *Georgetown Medical School, Washington, D. C.* In 1933 Chase and Lewis found that the rate of absorption of dl-methionine from the gastrointestinal tract of the white rat was 53 mg. per 100 gm. of body weight per hour. The method they employed to estimate the methionine remaining in the G. I. tract was the determination of amino nitrogen. The present work details the results of feeding both dl- and l-methionine to white rats and measuring the rate of absorption by two methods. First, the methionine remaining in the G. I. tract was determined by a modified McCarthy-Sullivan method and second, the total sulfur content of the tract was determined. By the colori-

metric method the rates of absorption were 40.0 mg. and 36.8 mg. per 100 gm. per hour for dl- and l-methionine respectively. From the determination of the total sulfur the calculated rates were 39.1 mg. and 35.4 mg. per 100 gm. per hour for dl- and l-methionine respectively. Both methionine and homocysteine were found in the livers of the rats fed methionine, there was no increase in the cystine or glutathione contents of the liver.

Manganese and thiamine in the diet of mother rats and body temperature control in the young. ROBERT M. HILL and DORSEY E. HOLTKAMP (by invitation). *Department of Biochemistry, University of Colorado Medical Center.* A recent paper from this laboratory (Am. J. Physiol. **149,** 650–656, 1947) presented evidence that nutritional factors in the diet of the mother may modify the time of development of body-temperature control against cold in young white rats. We can now report that the manganous ion is a promoting factor and that thiamine has an antagonistic action. The activity of each factor is most striking at low dietary levels of the other. In the first experiment Purina Fox Chow (700 gamma Mn and 30 gamma thiamin per day per mother rat) was used without supplement. The mothers were on this diet from the time of breeding until after the young were 18 days old. At that time the young were exposed in individual cages in the cold room at 3–6°C. No control of body-temperature was exhibited by these animals. In the second experiment 1000 gamma Mn was added to the diet per mother rat per day. At 18 days of age, one-third of the young had acquired almost complete body-temperature control against cold. In the third experiment, in addition to 1000 gamma Mn, 100 gamma thiamin was added per mother rat per day. On the 18th day of life none of the young of this group showed complete body-temperature control against cold, though most of them had developed some degree of resistance. This work was repeated using powdered whole milk as the basic diet. Similar results were obtained.

Stereochemistry of allopregnanetriol-3/16/20 of mare's urine. H. HIRSCHMANN, FRIEDA B. HIRSCHMANN (by invitation), and MARGARET A. DAUS (by invitation). *Department of Medicine, Western Reserve University and the Lakeside Hospital, Cleveland, Ohio.* Reduction with platinum in alcohol of the known 20-acetoxyallogregnanedione-3,16 derived from the allopregnanetriol-3,16,20 (I) of mare's pregnancy urine yields 20-acetoxyallopregnanol-3(β)-one-16 (m.p. 191.5–193°) which can be acetylated to a diacetate (II) (m.p. 184–186°). The same compound has been prepared by a second route. Partial solvolysis of the triacetate of I furnishes two diacetates melting at 165–166° and at 115–116.5°. The lower melting of these on oxidation with chromic acid is converted into II. Reduction of II with platinum in acetic acid or with nickel in alcohol followed by acetylation yielded a triacetate (III) identical with that prepared from pseudotigogenin diacetate by the method of Marker. It follows that triol I has erroneously been assigned the α configuration at C-3 and that like tigogenin it is a β-3-hydroxysteroid. Compound III has a higher rotation ($[\alpha]_D = +46°$) than the 20-epimeric triacetate ($[\alpha]_D = -25°$) prepared from tigogenin by means of persulfuric acid. The 16-acetoxyl group of the triacetate of I is solvolyzed faster than that of III which indicates that the 16-hydroxyl group of I is *trans* to the side chain at C-17.

2-Aminopurine as a purine antagonist. GEORGE H. HITCHINGS, GERTRUDE B. ELION (by invitation) and HENRY VANDERWERFF (by invitation). *The Wellcome Research Laboratories.* The relative effectiveness of 2-aminopurine as an inhibitor of the growth and acid-production of *Lactobacillus casei* depends on the nutrilites present and the duration of the experiment. This adenine isomer is an active inhibitor when folic acid is present but relatively ineffective when thymine is substituted for folic acid. In a medium containing 2-aminopurine and folic acid, the inhibition of growth can be relieved by the addition of adenine or folic acid. When the period of observation is extended from the usual 72 hours to 13 days the behavior of 2-aminopurine is found to be distinctive. With certain purine antagonists such as 5-amino-7-hydroxy-l-v-triazolo[d]pyrimidine acid production is continuous with a slight inflection in the curve at about the third day. With 2-aminopurine, however, acid production virtually ceases at the third day though the organisms remain viable. The production of acid by *L. casei* may be regarded as occurring in 2 phases, one of which may be completely inhibited by 2-aminopurine.

Coupled oxidations in enzymatically oxidized linoleic acid. R. T. HOLMAN (introduced by P. B. PEARSON). *Medicinska Nobelinstitutet, Stockholm and Dept. of Biochemistry and Nutrition, A & M College of Texas, College Station.* Using homogeneous crystalline lipoxidase enzyme and linoleic acid substrate at pH 9 the destruction of carotene and bixin added to the system has been studied. It has been found that in this homogeneous lipoxidase assay system used by Theorell, et al., the carotenoid destroyed is proportional to time and to enzyme concentration. Carotenoid oxidized is also proportional to the amount of diene conjugated (peroxide formed). Thus, under these conditions and in the range of concentrations studied, the disappearance of carotenoid is a valid measure of the enzyme activity and can be used as an assay. The oxidative destruction of carotene or bixin in linoleic acid oxidized by lipoxidase is proportional to its own concentration. The amount of diene conjugated decreases as carotenoid concen-

tration in the reaction mixtures is increased. It thus appears that the oxidation of carotenoid occurs as the result of the interruption of the chain linoleic acid oxidation reaction by carotenoid molecules. The number of linoleic acid molecules whose conjugation was inhibited by the oxidation of one molecule of carotenoid was found to be approximately 43 for carotene and 26 for bixin.

The kinetics of inhibitor action in a carrier-linked system. B. L. HORECKER and J. N. STANNARD (by invitation). *Laboratory of Physical Biology, National Institute of Health, Bethesda, Maryland.* Warburg and others have shown that the inhibition of respiration by cytochrome oxidase inhibitors increases when substrate is added, and have interpreted these results in terms of the "saturation" concept. Difficulties have arisen in the application of this concept. An alternative explanation is offered based on the kinetics of carrier-linked systems. From a consideration of the steady state, Equation 1 is derived to account for the effect of inhibitor on the overall reaction,

$$(1) \quad \text{Per cent inhibition} = \frac{1 - P}{1 + RP} \times 100$$

where R is the ratio of oxidase to dehydrogenase activity and P the fraction of oxidase activity remaining in the presence of inhibitor. The equation was tested by a study of the effect of azide on the succinoxidase system. The activities of cytochrome oxidase and succinic dehydrogenase were measured independently by spectrophotometric observation of the rates of oxidation or reduction of cytochrome c. The ratio R was calculated from the first order reaction velocity constants. P was calculated from the mass law expression for the dissociation of the cytochrome oxidase-hydrazoic acid complex, with dissociation constant of 7×10^{-7}. The O_2 uptake of the succinoxidase system was measured manometrically under the same conditions of O_2 tension, temperature and substrate concentration as in the spectrophotometric experiments. The observed inhibition of the O_2 uptake was found to be in agreement with Equation 1. Even at optimal substrate concentrations the overall reaction was inhibited considerably less than the oxidase. The equation accounts for increased inhibition with increase in substrate concentration.

Studies on cerebrosides. SIE HSIEN-GIEH (by invitation), C. E. FAUST (by invitation) and HAROLD H. WILLIAMS. *Department of Biochemistry, Cornell University, Ithaca, N. Y.* Previous studies have shown that cerebrosides, although prominent constituents of brain and nervous tissues, are present in other organs and tissues, particularly the muscles. Furthermore, they have been shown to have a common structure consisting of the nitrogenous base, sphingosine, the hexose, galactose, and fatty acid; variations in the type of cerebroside being dependent on the particular fatty acid present. Recent work, however, has demonstrated that another type of cerebroside may occur, especially in the spleen in Gaucher's disease. This type is characterized by the presence of a different hexose in the molecule, namely, glucose. In view of these observations, an investigation of the type of cerebroside (with respect to the hexose present) present in muscle and other tissues was undertaken. Cerebrosides were isolated from beef and rat brain, beef cardiac muscle, chicken breast muscle and egg. The hexose present was identified by chemical and physical methods and confirmed by specific microbiological fermentation procedures which will be discussed. In all cases, galactose was found to be the only hexose present.

Phosphorylation of glucose due to a coupled oxidation-reduction between α-ketoglutaric acid and oxalacetic acid. F. EDMUND HUNTER, JR. *Department of Pharmacology, Washington University School of Medicine, Saint Louis, Missouri.* Earlier work, J. Biol. Chem. **159**, 295 (1945), indicated that washed kidney tissue particles catalyzed the following reaction anaerobically: α-ketoglutarate + oxalacetate → succinate + CO_2 + malate. In attempting to explain the stimulating effect of this reaction on the conversion of acetoacetate to citrate, it was suggested that this anaerobic reaction, like the aerobic oxidation of α-ketoglutarate, could generate high energy phosphate bonds. The difference between the oxidation-reduction potentials of the oxalacetate-malate system and the α-ketoglutarate-succinate system is sufficient to permit such a reaction. Investigation reveals that this reaction does result in the creation of high energy phosphate which can be transferred via the adenylate system to glucose under certain conditions. Liver particles are more active than kidney. A homogenate prepared with phosphate-Ringer solution is centrifuged at high speed and the precipitate washed three times with phosphate Ringer. When the reaction mixture consists of tissue, oxalacetate, α-ketoglutarate, phosphate, Mg^{++}, DPN, NaF, adenylate, and glucose, very little glucose is phosphorylated, but the adenylate is converted to ATP. This is best seen with more than catalytic amounts of adenylate. When purified hexokinase is added with catalytic amounts of adenylate, inorganic phosphate disappears and glucose-6-phosphate is formed. Very little phosphorylation occurs when either oxalacetate or α-ketoglutarate is omitted. The pyruvate-lactate system, although of similar potential, will not replace oxalacetate-malate. These experiments represent a step toward direct demonstration of the points of origin of the three high energy phosphate bonds which result from aerobic oxidation of α-ketoglutarate to succinate.

The determination of various amines by proton-exchanges with ethyl eosin in non-aqueous solvents. J. LOGAN IRVIN and ELINOR MOORE IRVIN (by invitation). *Department of Physiological Chemistry, The Johns Hopkins University, School of Medicine, Baltimore.* Various nitrogenous compounds of biochemical interest can be determined in micro-quantities by means of proton-exchanges in non-aqueous solvents. For example, 0.1 μg. of pamaquine can be determined in 10 ml. of ethylene dichloride by measurement of the fluorescence of the anion of ethyl eosin which results from the transfer of a proton from ethyl eosin (practically non-fluorescent) to a proton-acceptor group in pamaquine. Of various fluorescein derivatives eosin and ethyl eosin are especially satisfactory because their anions have absorption bands of favorable wavelengths for excitation by the intense 545 mμ "line" of the mercury emission spectrum. Ethyl eosin is preferable to eosin inasmuch as the esterified carboxyl group of the former does not compete with the desired fluorescence-yielding proton-exchange. In contrast to water, which exerts a leveling effect through its amphoteric property, ethylene dichloride and similar solvents are inert and do not compete with the desired reaction through donation or acceptance of protons. The method has been applied in the determination of various amines such as certain antimalarial compounds, benzedrine, phenylethylamine, and histamine. Application to the latter two compounds suggests use of the method, in conjunction with specific amino acid decarboxylases, for micro determination of certain amino acids. Partial separation of primary amines from secondary and tertiary amines can be effected by reaction with a large excess of salicylaldehyde in alkaline aqueous solution. Upon extraction of the solution with ethylene dichloride, the salicylidine-imine, is retained in the alkaline aqueous solution.

Physical dependence liability of drugs of the methadon series and of 6-methyldihydromorphine. HARRIS ISBELL (by invitation) and A. J. EISENMAN. *Research Department, U. S. Public Health Service Hospital, Lexington, Kentucky.* Average doses of 21 mgm. of racemic methadon 6-dimethylamino-4-4-diphenyl-3-heptanone), 11 mgm. of levo-methadon, 90 mgm. of dextro-methadon, 84 mgm. of racemic methadol (6-dimethylamino-4-4-diphenyl-3-heptanone) 78 mgm. of racemic isomethadon (5-dimethylamino-4-4-diphenyl-3-hexanone) and 83 mgm. of 6- methyldihydromorphine were administered to 10 men, at the 28th to 32nd hour of abstinence from morphine. Levo-methadon and racemic methadon reduced the intensity of the abstinence more than did 30 mgm. of morphine. Dextromethadon and methadol had no effect. Isomethadon reduced the intensity of abstinence as much as did 30 mgm. of morphine. 6-Methyldihydromorphine had only a small and transient effect.

1 mgm. of levo-methadon was substituted for each 8–10 mgm. of the stabilization dose of morphine in 7 subjects who were addicted to morphine without the appearance of signs of abstinence. Following withdrawal of the levo-methadon after 14 days substitution, a definite abstinence syndrome ensued, which was similar to the syndrome seen after withdrawal of racemic methadon. Following the substitution of 1 mgm. of isomethadon for each 1.5 mgm. of the stabilization dose of morphine, signs of mild abstinence appeared in 2 of 5 cases. When the isomethadon was withdrawn after 14 days substitution, an abstinence syndrome, very similar to the morphine abstinence syndrome, became evident 12 hours after the last dose was administered.

Availability of dl-lanthionine for the promotion of growth when added to a cystine-deficient diet. D. BREESE JONES, ALVIN CALDWELL (by invitation), and MILLARD J. HORN (by invitation). *From the Bureau of Human Nutrition and Home Economics, Agricultural Research Administration, United States Department of Agriculture, Washington.* Lanthionine, the thioether amino acid isolated from various sodium carbonate treated proteins was obtained in two forms, the meso and racemic (Horn, Jones, and Ringel. J. Biol. Chem., **144,** 87 [1942]: **144,** 93 [1942]). It was later shown that the internally compensated mesolanthionine can not serve in lieu of cystine in the diet of the rat (Jones, Divine, and Horn. J. Biol. Chem., **146,** 571 [1942]). Inasmuch as rats are unable to utilize D-cystine for growth, their failure to grow on a cystine deficient diet supplemented with mesolanthionine indicates that they are unable to cleave the thioether amino acid or that cleavage occurs with the formation of the unutilizable D-cysteine instead of L-cysteine. It therefore seemed possible that L-lanthionine can be utilized for growth. Feeding experiments have now shown that racemic lanthionine can replace cystine and methionine in the diet. Young albino rats fed a low protein (casein) basal diet deficient in cystine but adequately supplied with the non-protein dietary essential factors declined in weight rapidly. For comparison with the behavior of rats given racemic lanthionine, L-cystine and DL-methionine were also used. Addition of 0.3 gm. of L-cystine or 0.37 gm. of DL-methionine per 100 gm. of basal diet caused immediate resumption of growth. Addition of DL-lanthionine likewise resulted in corresponding growth resumption. An immediate decline occurred when the lanthionine was omitted from the basal diet, and growth again was resumed when the lanthionine was added.

Citrate formation from oxalacetate. GEORGE KALNITSKY (introduced by H. A. MATTILL).

Department of Biochemistry, State University of Iowa, Iowa City. Rabbit kidney cortex homogenates form citrate from oxalacetate. Phosphate and bicarbonate-CO_2 buffers were most suitable. Borate and veronal buffers inhibited at higher (0.04M) concentrations. Addition of inorganic phosphate to homogenates resulted in slightly increased citrate formation. This was not due to buffering effect of the phosphate. Active cell-free preparations were obtained by centrifuging homogenates (1:2 dilution) for 4–5 minutes at approximately 3000 rpm. $Q_{\text{citrate formation}}$ was 6.9. The optimum pH was 7.3–7.9 with the peak at 7.5–7.6. Optimum substrate concentration was 0.02M (100 uM). Formation of citrate was proportional to enzyme concentration up to 1.5 ml. of enzyme solution in a total volume of 5 ml. Addition of pyruvate or acetoacetate did not appreciably increase citrate formation. An atmosphere of air or O_2 was equally effective; anaerobically, under N_2, 50% less citrate was formed. Dialysis for 15 to 20 minutes resulted in a marked decrease (66%) in citrate formation. Most of this activity was restored on addition of inorganic phosphate. The necessity of inorganic phosphate for citrate formation from oxalacetate might be interpreted as evidence for (1) the formation of a phosphorylated intermediate, possibly phosphocitrate, or (2) the combination of oxalacetate with a two carbon compound, according to the following reactions: oxalacetate $\rightarrow$ pyruvate $\xrightarrow{PO_4}$ 2-c-compound; oxalacetate + 2-c-compound $\rightarrow$ citrate. After dialysis for 50 minutes against distilled water the preparation was inactivated and could not be reactivated by addition of inorganic phosphate, boiled kidney juice and adenosinetriphosphate.

Reactions between acetate, acetyl phosphate and the adenylic acid system in tissue and bacterial extracts. NATHAN O. KAPLAN (by invitation) and FRITZ LIPMANN. *Biochemical Research Laby., Massachusetts General Hospital, and the Department of Biological Chemistry, Harvard Medical School, Boston.* Phosphate exchange between acetyl phosphate and the adenylic acid system had readily been observed with various bacterial extracts, either by direct reaction or by using glucose as the ultimate P-acceptor. A transfer of acetyl-P to glucose has now been observed also in pigeon liver extracts. In aged acetone powder extracts, a fixation of from 18 to 55% of the added acetyl P occurred, using fluoride and citrate as inhibitors of interfering enzymes. Higher values of 36–55% were obtained with added ADP. On omitting glucose, no fixation took place. This phosphate transfer from acetyl phosphate to the adenylic acid system does not require coenzyme A, although Co A is necessary for acetyl transfer from acetate plus ATP. Recently, in studies with E. coli extracts acetate plus ATP were found to yield large amounts of acetyl phosphate without addition of an acetate acceptor. The compound formed seems to differ, however, from synthetic monoacetyl phosphate by being quite resistant to the action of an earlier described specific muscle acetyl phosphatase. Efforts are underway to test for the existence of two acetyl-P-derivatives by isolation of the product of the reaction between ATP and acetate.

Isolation and assay of an anti-secretory substance from duodenal mucosa. J. KATZ (by invitation), ROBERT L. DRYER (by invitation), W. D. PAUL (by invitation), and J. I. ROUTH. *Departments of Biochemistry and Internal Medicine, College of Medicine, State University of Iowa, Iowa City.* Extracts have been prepared from lyophilized de-fatted duodenal mucosa of hogs according to the method of Kim and Lim. Various methods have been applied to concentrate and purify a substance from these extracts which actively inhibits gastric secretion and gastric ulceration. The concentrates were tested by intraperitoneal injection of 1 cc. into rats with ligated pylorus. The animals were prepared by fasting 72 hrs. with free access to water. Pyloric ligation was performed under light ether anesthesia, and the extracts were injected immediately following ligation. After 6 hr. the stomachs were excised and the volume of the contents was measured and compared with that from control rats, with ligated pylorus, injected with 1 cc. of normal saline. Free acidity, total acidity, pH, and peptic activity was determined in all samples. The interior of each stomach was examined with a dissecting microscope. Our results indicate that the material isolated from these extracts possesses an inhibitory effect on gastric secretion, an effect which we are attempting to utilize as an assay method. The anti-ulcer activity of this substance is also under study. Attempts are being made to correlate the anti-secretory and anti-ulcer effects with the physical and chemical properties of the active agent.

Application of the isotopic derivative method of analysis to protein hydrolysates. ALBERT S. KESTON (by invitation), SIDNEY UDENFRIEND (by invitation), and R. KEITH CANNAN. *Department of Chemistry, New York University College of Medicine, New York.* The isotopic derivative method, recently reported (Keston, Udenfriend, and Cannan, J.A.C.S., 68, 1390 (1946)), has been modified and further applied. Various techniques for separating the homologous derivatives were studied to separate different carriers as well as to aid in their purification. Distribution coefficients of many p-iodophenyl sulfonyl (pipsyl) amino acids between various solvent pairs were determined. The relative distribution coefficients of the pipsyl amino acids between chloroform and water are

almost the same as those of the acetyl derivatives (Synge, Biochem. J. 33, 1913 (1939)) although the two substituent groups differ greatly in size. The distribution coefficients were used to develop countercurrent distribution procedures for the separation of the different derivatives, making it possible to add several carriers to a protein hydrolysate which has been treated with isotopic pipsylchloride, and to obtain analyses for several amino acids on a single aliquot. By using several different techniques for purification of individual carriers purity is more readily achieved. Countercurrent solvent extractions, recrystallizations, and differential adsorption on norite were all used to achieve purification of the isolated carriers. Recoveries of glycine, alanine, and proline from known mixtures of amino acids have been highly satisfactory. When the amino acid being estimated was omitted from the mixture none was found by the method. This demonstrated the separability of homologous isotopic impurities from the carriers. Analytical values have been obtained for glycine, alanine, and proline in β-lactoglobulin, human hemoglobin, rabbit aldolase, and rabbit phosphoglyceraldehyde dehydrogenase, using samples of protein of the order of 1 mg.

Quantitative analysis of protein hydrolysates on paper chromatograms by means of the isotopic derivative method. ALBERT S. KESTON (by invitation), SIDNEY UDENFRIEND (by invitation), and MILTON LEVY. *Department of Chemistry, New York University College of Medicine, New York.* The isotopic derivatives obtained in the method of analysis described by Keston, Udenfriend, and Cannan (J.A.C.S., 68, 1390 (1946)) are suitably separated by paper chromatography. The combination offers a method applicable to the estimation of amino acids in microgram quantities of proteins. A sample of derivatives, labelled with p-I^{131}-$C_6H_4SO_2Cl$ (I^{131}-pipsylchloride) is placed as a transverse line on a paper strip and the chromatogram developed with n-pentanol saturated with 2 N NH_3. Different derivatives are located as radioactive bands with a G.M. counter or radioautography, and are initially identified from retardation factors. The counts in a resolved band divided by C_r, counts per mole of substance made with the same reagent, equals the moles of substance. Identity and purity of a band is established by eluting, adding corresponding non-isotopic derivative as carrier, and demonstrating unchanged isotope concentration on purification. Estimations may be made independent of complete resolution of bands by adding indicators (either unlabelled derivative or derivative labelled with a second isotope) before chromatography. Estimations then depend on isotope ratios in any pure sample. To determine an amino acid, its S^{35}-labelled pipsyl derivative (B counts) is added as indicator to the derivatives obtained from a hydrolysate and I^{131}-pipsylchloride. Where the ratio of I^{131}/S^{35}, (R), in successive strips is constant it can be used to calculate the amount of amino acid originally present; moles of amino acid = RB/C. R is readily calculated from measurements made with and without an Al filter.

A new adenosinetriphosphatase of muscle. W. WAYNE KIELLEY (introduced by OTTO MEYERHOF). *Department of Physiological Chemistry, School of Medicine, University of Pennsylvania.* A highly unstable ATP-ase has been isolated from muscle by extraction with dilute alkaline salt solution, repeated fractionation with 0.3 to 0.4 saturated $(NH_4)_2SO_4$ and high speed centrifugation (18000 r.p.m.) This enzyme which splits ATP to ADP is activated by Mg, inhibited by Ca, and has an optimum activity around pH 7.4. In all these respects it differs from the myosin ATP-ase. It is present in fresh muscle in about the same amount as the myosin ATP-ase and can be brought to the same degree of absolute activity (purity) as the latter. In the presence of some ampholytes the activation and pH optimum of myosin may be shifted toward the properties of this new preparation. The relation between the two enzymes will be discussed.

Effect of carbon dioxide concentration on the changes in brain metabolites accompanying convulsions. J. RAYMOND KLEIN and JAMES A. BAIN (by invitation). *Depts. of Psychiatry, Biological Chemistry, and Pharmacology, University of Illinois College of Medicine, Illinois Neuropsychiatric Institute, Chicago.* Convulsions in cats breathing a mixture containing 15% carbon dioxide and 85% oxygen were found to be accompanied by an increase in brain lactate, a decrease in glucose, and no obvious decrease in concentration of adenosine polyphosphates or phosphocreatine. The absolute increase in brain lactate was less than that found in convulsed animals breathing room air and the level attained was about the same as in unconvulsed animals breathing room air. The lack of change in labile phosphates in the convulsed animals breathing the carbon dioxide mixture contrasts with the considerable decreases occurring in convulsed animals breathing room air.

Selenium tetraglutathione. HARLAN L. KLUG (by invitation) GEORGE P. LAMPSON (by invitation) and ALVIN L. MOXON. *Chemistry Department South Dakota Agricultural Experiment Station, South Dakota State College, Brookings, South Dakota.* Sodium selenite (subcutaneously injected) has been shown to decrease liver and blood reduced glutathione (GSH) values in rats in this laboratory. These results suggested that selenite had combined with GSH in the tissues; therefore an investigation of the reaction between selenite and GSH was initiated. The compound was prepared by mixing

aqueous solutions of .01 mole GSH and .0025 mole selenious acid; ethanol was added until a white precipitate formed; the product was cooled overnight on ice, filtered, washed with ethanol, and dried in a vacuum desiccator. Analysis showed a sulfur to selenium ratio of 4:1. The crystalline compound is white, slightly soluble in cold H_2O, moderately soluble in hot H_2O, and is stable in an acid solution. Decomposition occurs in an alkaline solution with liberation of elemental selenium, and also upon heating the compound to 140°C.

Tests on an aqueous solution show neither free SH groups nor excess selenious acid, but reduction by zinc dust in acid solution liberates elemental selenium and SH groups which can be recovered in theoretical amounts when titrated with KIO_3. Tests show only a trace of disulfide sulfur, which indicates that practically all the sulphur is bound to selenium. These tests indicate further that the product formed is a selenium-tetraglutathione with one Se atom bound to the four S atoms. The compound is significant in selenium toxicity studies as it may assist in explaining the detoxication effect of SH groups on selenium. Toxicity studies show that the selenium in this compound is markedly less toxic than selenium in sodium selenite, which indicates that the combining of selenite with GSH converts the selenium to a less toxic form.

Studies on the micro molecular distillation of blood lipids. ALFRED E. KOEHLER, ELSIE HILL (by invitation) and FRANK FEARNEY (by invitation). *Santa Barbara Cottage Hospital and The Sansum Clinic Research Foundation, Santa Barbara, California.* Further studies of the distillation of serum lipids from a film at .003 micron Hg at comparatively low temperatures were made. It was found that 88°C. was the most suitable temperature for the separation of cholesterol from its esters. This separation agreed closely with the digitonin method in normal sera but when the esters were low the free cholesterol was about 10 % lower than that by the digitonin separation. This indicates the presence of a substance that gives the Liebermann-Burchard color which is precipitated by digitonin but is not distilled under conditions that completely distill free cholesterol. Fatty acids can readily be distilled, partially even at room temperature, 50% at 35°C., 90% at 128°C. After phospholipid and free cholesterol separation by precipitation, the fatty acid glycerides can be partly separated from the cholesterol esters by distillation at 100°C. or lower inasmuch as no natural cholesterol esters distill below 100°C. Approximately 50% of the blood fats distill at 88°C. Fractionation of the combined fatty acids of the Bloor filtrate, as well as the fats, shows a fairly uniform composition of different sera in health and a variety of disease conditions. Some notable exceptions were found however, especially in some instances of diabetic ketosis.

DPN pyrophosphatase. ARTHUR KORNBERG and OLOV LINDBERG (introduced by CARL F. CORI). *Department of Biological Chemistry, Washington University School of Medicine, St. Louis.* Enzymes which split diphosphopyridine nucleotide (DPN) are found in microorganisms, in plant and in animal tissues. The mode of action of these enzymes is not well understood. Handler and Klein (J. Biol. Chem. **143:** 49, 1942) reported that in animal tissues the nicotinamide-ribose linkage is the primary and principal site of cleavage. Heiwinkel (Arkiv. Kemi. Mineral. o. Geol. **13A:** 19, 1939) observed the formation of adenylic acid when DPN was incubated with sweet almond press juice. We have observed that washed rabbit kidney particles convert added DPN to adenylic acid and a product which has been identified as nicotinamide ribose phosphate. In the presence of an oxidizable substrate (i.e. glutamate) as a source of phosphate bond energy, adenosine triphosphate is formed in place of adenylic acid. These and other observations indicate that the pyrophosphate bond is the primary site of DPN cleavage in these preparations.

DPN is split more rapidly in a respiring system (i.e. with added glutamate) than in a non-respiring one. This fact may be explained by the more rapid rate of splitting of the reduced form of DPN as compared with the oxidized form, as measured under anaerobic conditions.

The determination of the biological indices of gluten and gluten fortified with lysine in normal and protein-deficient patients. DONALD D. KOZOLL (by invitation), WILLIAM S. HOFFMAN, GORDON MCNEIL (by invitation) and HANS POPPER (by invitation). *From the Hektoen Institute for Medical Research of the Cook County Hospital, Chicago, Illinois.* The method of Allison was applied to 10 human subjects in whom gluten (G) and gluten fortified with 4% lysine (GL) were administered orally. Seven patients were protein-deficient; 3 normals were studied for comparison. All patients were kept 3 to 9 days on a protein-free diet for the determination of the endogenous nitrogen excretion. Thereafter, 3 protein deficient subjects were fed for 5 day periods progressingly increasing levels of intake of *G* and *GL*. In the remaining 7 subjects a simplified procedure was utilized consisting of 10 day periods during which equivalent quantities of *G* or *GL* calculated to barely approach equilibrium were given. In 3 subjects an additional 10 day period was employed wherein the patient returned to *G*.

The biological index (K) for *G* varied from 0.70 to 1.03; for *GL* this value varied between 0.82 and 1.22. The calculated quantities of nitrogen required for equilibrium for *G* varied between 49 and 109

mg. per kilo per day, whereas, for *GL* it ranged between 39 and 98 mg. per kilo per day. No differences were discernable between normal and protein-deficient subjects. Where patients were returned to *G*, results were similar to those initially obtained with *G*. Eight subjects were submitted for statistical analysis. The average *K* value for *G* was 0.63 and for *GL* was 0.78. The average nitrogen requirement for nitrogen equilibrium for *G* was 83 mg. per kilo per day and for *GL* 68 mg. per kilo per day. The conclusions are drawn that the determination of the biological index of Allison is applicable to the study of an oral protein in the human protein deficient subject. Although gluten proved to be a fairly complete protein, gluten fortified with 4% lysine had a higher biological value.

Immunochemical studies on purified d-glyceraldehyde-3-phosphate dehydrogenase from yeast and rabbit muscle. EDWIN G. KREBS (by invitation) and VICTOR A. NAJJAR. *Department of Biological Chemistry, Washington University School of Medicine.* Antibodies for d-glyceraldehyde-3-phosphate dehydrogenase from yeast have been produced in rabbits. The enzyme used for the antigen was obtained from baker's yeast by a simplification of the procedure reported by Warburg and Christian and was 85% pure by electrophoretic measurement. Several recrystallizations led to electrophoretically homogeneous enzymes, but this was not used for immunization. Rabbits were given a series of subcutaneous injections of the antigen and developed precipitating antibodies. Precipitin tests with undiluted serum were positive to a 1:10,000 dilution of the 4 per cent antigen solution. The activity of the yeast enzyme was inhibited 80 per cent by a 1:30 dilution of the antiserum in the reaction mixture. No inhibition of crystalline muscle d-glyceraldehyde-3-phosphate dehydrogenase was noted under the same conditions. Further studies are being undertaken to determine whether antibodies to the yeast enzyme produced in a different animal will inhibit the rabbit enzyme.

The crystalline yeast and rabbit enzyme were compared in other properties. Kinetically the specific activities are of the same magnitude, and both enzymes are activated by cysteine. This latter fact was not reported by Warburg for the yeast enzyme. Taylor et al. (in press) have found that the muscle enzyme crystallizes with DPN as an essential component of the crystalline structure. No DPN has been detected in the crystalline yeast enzyme. At pH 7.3 the electrophoretic mobility of the yeast enzyme is much greater than that of the muscle enzyme.

Nutritional studies on the cat. W. A. KREHL and I. D. WELT (by invitation). *Yale Nutrition Laboratory, Department of Physiological Chemistry, Yale University.* No adequate data are available concerning the nutritional requirements of the cat, an animal which may be profitably used for nutritional investigations. The present communication represents a successful attempt to maintain the growth of kittens to maturity upon a completely "synthetic" diet. In our experience cats would not eat the usual synthetic rat diet. A diet modeled after the composition of solids in cows' milk resulted in maintenance, but was inadequate for growth. A ration based upon the composition of the solids of bitch's milk, an animal with a rapid growth rate similar to that of the cat, was then devised. All recognized vitamins of the B-complex, as well as synthetic vitamin K, were added to this basal ration. A concentrate containing appropriate amounts of vitamins A, D, and E was administered weekly by mouth.

Three female litter-mate kittens, one litter-mate male, and one non-litter-mate male were placed upon this ration. It has proved to be eminently satisfactory for growth beyond maturity to a degree comparable to that reported in the literature for kittens fed the usual "house" diet. The adequacy of this diet for normal growth was confirmed as follows: One male and two female kittens were placed on a similar diet, to which 4% of whole liver powder was added at the expense of lard. No significant difference could be detected between the growth rates of the two groups of animals. Blood studies were also carried out and will be reported.

The availability of amino acids in some foods. K. A. KUIKEN (by invitation) and CARL M. LYMAN. *Department of Biochemistry and Nutrition, A. and M. College of Texas, College Station.* The extent to which the individual amino acids in a number of foodstuffs are available for use by the rat was studied by determining the 10 essential amino acids in the food and in the feces. Young rats were fed rations in which practically all of the protein was supplied (at a 10% level) by a single foodstuff. The experimental period was followed by a control period in which a ration containing 4% whole egg protein was fed. The data obtained from the control period were used to correct the apparent availabilities for the amino acids normally present in the feces as a result of metabolic processes and bacterial action.

The results showed that the individual amino acids in a given foodstuff are not always equally available. For example, the value obtained for the availability of arginine in cottonseed flour was 93.4% while the value for the availability of lysine in the same sample was 64.5%. In the case of meat all 10 amino acids were 100% available. The corresponding values for wheat ranged from 92.2 to 98.8% and for peanut flour from 94.8 to 99.5%.

The fatty acid oxidase complex of rat liver an

intracellular structures. ALBERT L. LEHNINGER and EUGENE P. KENNEDY (by invitation). *Departments of Surgery and Biochemistry, University of Chicago, Chicago.* In a recent investigation (Lehninger and Kennedy, J. Biol. Chem., submitted) the requirements for activity of the fatty acid oxidase system have been more completely defined. Saline-washed particulate matter of rat liver after treatment with water requires the presence of adenine nucleotide, Mg^{++}, inorganic phosphate, neutral salts such KCl (or certain non-electrolytes such as sucrose), cytochrome-c and catalytic amounts of l-malate or oxalacetate for restoration of ability to oxodize octanoate to acetoacetate. The function of the neutral salt is to cause transformation of the enzyme complex from an inactive "dispersed" condition to the enzymatically active, "flocculated" state. This behavior of the enzyme complex suggested possible identity with the "mitochondria" or "large granules", separable from liver extracts by differential centrifugation, which are known to "agglutinate" in isotonic saline. Also, the highly organized nature of the enzyme complex, which also catalyzes the reactions of the Krebs cycle and coupled phosphorylations, suggested that the particles preexist as such in the cell. Fractionation of the particulate matter of rat liver in 30% sucrose (Hogeboom, Schneider and Pallade, Proc. Soc. Exptl. Biol. Med., 65, 320 (1947)) revealed that the washed fraction designated by Hogeboom et al. as consisting of morphologically intact mitochondria, free of extraneous elements, contains essentially all of the fatty acid oxidase activity of rat liver as well as Krebs cycle activity.

Enzymatic oxidation of tyrosine and dihydroxyphenylalanine by melanoma extracts. AARON BUNSER LERNER (by invitation). THOMAS B. FITZPATRICK (by invitation), EVAN CALKINS (by invitation), and WILLIAM H. SUMMERSON. *Biochemistry Section, Medical Division, Army Chemical Center, Maryland.* The enzymes tyrosinase and dopa oxidase in melanoma tissue have been considered to be separate substances catalyzing the oxidation of tyrosine and dihydroxyphenylalanine (dopa) respectively. Experiments to be described indicate that the enzymatic oxidation of both of these substances by active fractions from mouse melanoma tissue can be accounted for in terms of a single enzyme. This enzyme appears to be actually a tyrosinase, with dopa, or a product of dopa oxidation, serving merely to catalyze the reaction in which tyrosine is converted to melanin.

Our evidence indicates that this enzyme, like plant tyrosinase, is a copper protein. Analysis of enzyme preparations for iron, cobalt, magnesium, manganese and copper revealed that only copper is present in significant amounts. When substances which combine with copper, e.g., sodium diethyl dithiocarbamate, phenylthiourea and others were added to enzyme preparations, the tyrosinase activity was inhibited. Enzymatic activity was restored by the addition of an excess of copper. Other metals did not produce this effect.

The effect of testosterone and methyl testosterone on guanidoacetic acid in blood and urine. BLAINE H. LEVEDAHL (by invitation) and LEO T. SAMUELS. *From the Department of Biochemistry, University of Utah School of Medicine, Salt Lake City, Utah.* With 3 male subjects maintained on a dose of 50 mg. of methyl testosterone per day, the blood and urine levels of creatine and guanidoacetic acid were found to increase sharply after an initial delay of approximately 30 days. Ten days after the cessation of methyl testosterone administration the creatine and quanidoacetic acid levels of both the blood and urine were found to be normal. Significant changes in the blood and urine creatinine levels were not observed during the test period. With 3 female subjects given 50 mg. of methyl testosterone per day, the rapid use in the creatine and guanidoacetic acid levels was found to occur approximately 12 days after the start of administration. Increases of 3 to 6 times the normal urine creatine and guanidoacetic acid levels as well as increases of 2 times the normal blood levels of creatine were observed in a period of 10 days. No significant changes in the blood and urine creatine levels were noted.

Testosterone propionate given by injection, implantation, or oral administration to four male subjects for 40 days showed only a decrease in the urine creatine level after approximately 25 days of administration. Methyl testosterone appears to increase formation of guanidoacetic acid as well as creatine and therefore is probably not primarily involved in transmethylation.

The effect of gonadotrophic stimulation on the cholesterol content of the immature rat ovary. LOUIS LEVIN and JOSEPH W. JAILER (by invitation). *Departments of Anatomy and of Obstetrics and Gynecology, College of Physicians and Surgeons, Columbia University, New York.* Bloch has shown that fed cholesterol is excreted in part as pregnandiol, the conversion presumably being by way of progesterone. It may be expected that during the ovarian secretory process, ovarian cholesterol is utilized for conversion to estrogen and/or progesterone. Accordingly, a comparison has been made of the cholesterol content of the immature rat ovary before and after treatment with pituitary gonadotrophin. Injection of this gonadotrophin causes a rapid loss of ovarian cholesterol (as much as 29% loss) followed by a gradual return toward the original level and a final attainment of levels far above the original. The loss of ovarian cholesterol is evident within 1 hour after a single injection of gonadotrophin and is maximal at 3 to 12

hours. Thereafter, cholesterol replenishment begins, the original level being nearly attained at 24 hours and being surpassed (double the original level) at 48 hours. The cholesterol accumulation continues at least until 100 hours after gonadotrophin administration. Due to the loss of cholesterol the concentration in the ovaries initially drops sharply. Later, during the phase of cholesterol accumulation, the concentration does not show any substantial increase because of the coincident increase in ovarian weight.

Human chorionic gonadotrophin, in adequate dosage, causes a loss of ovarian cholesterol similar to that caused by pituitary gonadotrophin. Such loss does not occur after injection of pituitary adrenotrophic hormone nor after exposure of the animal to cold, but these treatments do cause a loss of adrenal cholesterol.

Dibenzothiophene as a reagent for aldehydes, ketones and carbohydrates. VICTOR E. LEVINE and SIDNEY MERLIS (by invitation). *Department of Biological Chemistry and Nutrition, Creighton University School of Medicine, Omaha, Nebraska.* Dibenzothiophene condenses with carbohydrates and certain aldehydes and ketones in the presence of concentrated sulphuric acid with the formation of colored complexes. The reagent is 1% dibenzothiophene in alcohol. If the tested substance is in aqueous solution, the reagent is likely to precipitate. This precipitation does not interefere with the formation of a ring or zone of characteristic color, when reagent and solution are underlayed with sulphuric acid. Pentoses and pentose-yielding compounds (arabinose, xylose, ribose, fucose, rhamnose, gums, nucleic acid) yield a deep red ring. Hexoses and hexose-yielding compounds (glucose, fructose, sorbose, galactose, mannose, cellose, sucrose, maltose, melibiose, lactose, melezitose, raffinose, dextrin, starch, inulin; egg and serum albumin, casein; amygdalin, esculin) yield a blue ring changing to purple on shaking. A number of aldehydes and ketones form a pink ring. Acetaldehyde reacts, but acetone does not. p-Dimethylaminobenzaldehyde forms a purple ring. Formaldehyde yields an intense green color, specific and very sensitive, one gamma per milliliter being readily detectable. Formic acid does not interfere. Paraformaldehyde also reacts, as well as compounds which are condensation products of formaldehyde with ammonia (methenamine, methenamine mandelate) and with alcohols or thio alcohols (methylal, ethylal, propylal, djenkolic acid). The benzothiophene reaction detects formaldehyde in milk and also methyl alcohol after oxidation. It also serves to identify methyl alcohol in ethyl alcohol. Oxidation produces formaldehyde and acetaldehyde. The reagent in the presence of these two aldehydes forms an upper pink or reddish ring with acetaldehyde and a lower deep ring with formaldehyde.

The arrangement of amino acids in silk; an application of the isotopic derivative technique. MILTON LEVY and EVELYN SLOBODIANSKY (by invitation). *Department of Chemistry, New York University College of Medicine, New York.* Using p-$I^{131}C_6H_4SO_2Cl$ (pipsyl chloride) and the principle described by Keston, Udenfriend, and Cannan (J. A. C. S., **68**, 1390 (1946)), hydrolysates of silk fibroin, containing dipeptides, have been analysed for alanine, glycine, alanylglycine, glycylalanine, and glycylglycine. The hydrolysates were prepared by the action of 12 N HCl at 39° for 16 to 48 hours. The 48 hour hydrolysate contained, in per cent of N, 12% of glycine N, 9.5% of alanine N, 1.5% of glycylglycine N, 8% of glycylalanine N, and 25% of alanylglycine N. All the values were lower at 24 hours except glycylalanine which was unchanged. Total hydrolysis of the same silk fibroin gave 39% glycine N and 26% alanine N. Practically all of the alanine and about half of the glycine are accounted for in the 48 hour hydrolysate. The results are incompatible with a random arrangement of the amino acid residues in the protein.

The excretion of labeled calcium by normal and thyroparathyroidectomized rats. MAURICE V. L'HEUREUX (by invitation), WILBUR R. TWEEDY, and ELINOR M. ZORN (by invitation). *Department of Biological Chemistry, Loyola University School of Medicine, Chicago.* Young adult rats which were maintained on a stock diet containing 1.18% of calcium and 0.86% of phosphorus were used. Following the subcutaneous administration of 7 to 14 mgm. of labeled calcium (as $CaCl_2$) to 8 normal rats (av. wt. 247 gms.), 2.9% of the injected radiocalcium appeared in the urine within 6 hours. At the end of 24 hours, 4.1% of the administered radiocalcium was detected in the urine, and 8.9% in the feces and intestinal contents. In 2 other normal rats (av. wt. 240 gms.) the excretion of injected radio-calcium was determined daily for 8 days. At the end of this period 8.6% of the radiocalcium had been excreted in the urine and 18% in the feces.

Eight rats were thyroparathyroidectomized, and 1 to 39 days later injected with labeled calcium and sacrificed after 24 hours. 4 animals which showed terminal serum calcium values of 5.6 to 7.9 mgm. %, excreted 50 to 70% of the amount of radiocalcium excreted by their controls. The other 4 animals, whose terminal serum calcium values ranged from 8.3 to 10.4 mgm. %, excreted 101 to 117 % of the amount of radiocalcium excreted by their controls.

Adrenal cortical metabolites in human urine. SEYMOUR LIBERMAN (by invitation), DAVID K. FUKUSHIMA (by invitation), and KONRAD DOBRINER. *Sloan-Kettering Institute for Cancer Research.* Four new metabolites of adrenal cortical

steroids have been isolated from human urine. They are: (I) pregnanol-3α-dione-11,20; m.p. 174.5–5°; $[\alpha]_D^{22}$ = %111°; (II) androstanol-3α-dione-11,17; m.p. 175–175.5°; (III) etiocholanediol-3α,11β-one-17; m.p. 235–7°; and (IV) pregnanediol-3α,20α-one-11 (isolated as the diacetate, m.p. 231.5–3°; $[\alpha]_D^{24}$ = +56.7°.) These were identified by infrared spectroscopy. In each instance the structure was confirmed by comparison with authentic samples prepared by independent methods. (I) and (IV) upon oxidation with CrO_3 yielded pregnanetrione-3,11,20 identical with the known compound. The presence of the C_{11}-oxygen function in these compounds makes it almost certain that they are of adrenal origin. Since two of the products are C_{21} steroids, they can be presumed to arise from precursors such as corticosterone; the antecedents of the two C_{19} compounds cannot be so readily assigned.

There have been isolated from urine by us and others eight ketosteroids of unquestionable adrenal origin. In addition to the four reported above, pregnanediol - 3α,17α - one - 20, androstanediol - 3α,11β-one-17, etiocholanol-3α-dione-11,17, and Δ^5-pregnenediol-3β,17α-one-20 have also been isolated. Three transformation products related to these adrenal metabolites have been described. They are Δ^9-androstenol-3α-one-17, Δ^9-etiocholenol-3α-one-17 and 17a-methyl-Δ^5-D-homoandrostenediol-3β,17a (α)-one-17. The significance of these results for normal and abnormal steroid metabolism in man will be dealt with in the discussion.

New penicillin products for prolonged blood levels. Leo Loewe (by invitation) Albert Sobel, Oscar Gawron (by invitation) and Erna Alture-Werber (by invitation). *Departments of Medicine and Biochemistry, The Jewish Hospital of Brooklyn* 16, *New York.* A study was made of various preparations of penicillin in order to obtain sustained blood levels with a single subcutaneous or intramuscular injection. The following products gave levels for a period of 24 hours or more in rabbits. With similar amounts of penicillin (as the sodium salt) in aqueous solutions no measurable levels were obtained after 6 hours, by the method employed [J. Bact. 48, 599, (1944)].

1) Gelatin and penicillin salt mixtures treated with formaldehyde.
2) Penicillin salts dissolved or suspended in polyoxyalkalene derivative of sorbitan monolaurate.
3) The precipitate obtained by the treatment of (serum or milk) protein penicillin mixture with the salts of metal ions which form insoluble derivatives with both protein and penicillin. The metallic ions chosen were those which are nutritionally required. e.i., Fe, Cu.
4) Products of type 1) and 3) suspended in polyoxyalkalene derivative of sorbitan monolaurate.

On the origin of stercobilin in humans. Irving M. London (by invitation), Randolph West, David Shemin and D. Rittenberg. *From the Departments of Medicine and Biochemistry, College of Physicians and Surgeons, Columbia University, and the Presbyterian Hospital in the City of New York.* The feeding to humans of glycine labelled with N^{15} results in the incorporation of N^{15} in the heme of the red blood cells during their formation in the bone marrow. The labelled porphyrin remains in the red cell until the cell disintegrates. The destruction of the cells begins about 70 days after the formation of the cells and reaches a maximum at 128 days. The prophyrin is not reutilized for new hemoglobin synthesis but is excreted in large part as stercobilin. We have isolated crystalline stercobilin from the stools of subjects fed N^{15} labelled glycine for two days. The data indicate that not all the stercobilin results from the destruction of hemoglobin. For example, in one experiment the stercobilin isolated from the stools of the first eight days of the experiment contained 0.480 atom % excess N^{15}. During this period the cells being destroyed were formed before the administration of labelled glycine and therefore contained no labelled heme. Consequently one can conclude that a significant portion of normal stercobilin production is derived from sources other than hemoglobin. From a consideration of the N^{15} concentrations in the stercobilin and heme, it would appear that there is a source of stercobilin other than the known porphyrin-protein systems. Further work is in progress to delineate more clearly the relationship between hemoglobin destruction and stercobilin formation and to determine the other biological sources of bile pigment.

Fractionation of liver proteins. J. Murray Luck and A. Clark Griffin (by invitation). *Department of Chemistry, Stanford University, California.* A study has been made of the liver proteins of rats on standard diets with preliminary fractionation studies carried out on the livers of dogs and rabbits. The animals, anesthetized with nembutal, were viviperfused with Ringer-Locke solution until the livers were virtually blood-free. Fractionation of the proteins was then effected by successive extractions of the homogenized liver at pH 7 to 7.5 with sodium chloride solutions of varying molarity. The following fractions were obtained: albumin, globulin, ribonucleoprotein, desoxyribonucleoprotein, and final residue. Extraction of the ribonucleoprotein, assumed to be derived from the cytoplasm, could not be achieved by the use of 0.14 *M* sodium chloride (physiological saline). The studies, quantitative in character, include the livers of normal rats, livers in the pre-

cancerous state, and livers in which pronounced tumors had appeared. Azo dyes were used as the carcinogenic agents. All fractionations were carried out at 1°C. The values found for total liver ribose and total desoxyribose in the normal rat approximate those reported in the literature.

Studies on the agglutination of human and rat red cells by caster bean extracts. STEPHAN LUDEWIG and ALFRED CHANUTIN. *Biochemical Laboratory, University of Virginia.* It is well known that castor bean extracts will agglutinate red, white and tissue cells. An attempt has been made to determine the influence of a variety of physical and chemical changes which affect agglutination. Data will be given for human and rat red blood cells. Since rat cells are comparatively fragile it was necessary to determine conditions which would prevent hemolysis and this was done by adding small amounts of serum albumin. It was found that plasma, serum albumin, globulins, casein and other proteins affect the degree of agglutination. In addition, ionic strength, different halogens and pH influence this phenomenon. The red cells of rats poisoned with castor bean extracts behave similarly to those of control animals. The technique for the quantitative determinations of agglutination is critical and the procedures will be given.

Effect of vitamin B_6 on the utilization of D amino acids by lactic acid bacteria. CARL M. LYMAN and K. A. KUIKEN (by invitation). *Department of Biochemistry and Nutrition, A and M College of Texas, College Station.* It was found that the ability of *Lactobacillus arabinosus* to utilize the non-natural forms of the amino acids was radically altered when pyridoxamine was substituted for pyridoxine in the basal medium. For example, only negligible amounts of the D forms of isoleucine and leucine were utilized when the medium contained pyridoxine, but considerable amounts of these isomers were utilized when the medium contained pyridoxamine. The extent of utilization of D isoleucine in the presence of pyridoxamine was dependent on the concentration of leucine. Extensive utilization of D isoleucine occurred when the leucine level was 0.4 mg. per tube but this utilization was completely blocked by increasing the leucine level to 2 mg. per tube. Although *L. arabinosus* can partially utilize D glutamic acid the extent of this utilization did not appear to be dependent on the form or amount of vitamin B_6 in the medium. It was found that vitamin B_6 is not the only factor which modifies the ability of the lactic acid bacteria to utilize D amino acids. *Streptococcus faecalis* R did not utilize D methionine in a medium buffered with acetate, but substantial utilization occurred in a medium buffered with citrate. The significance of these findings in relation to the microbiological determination of amino acids is discussed.

Study of the oxidation of the labile methyl group of dietary methionine traced with C^{15}. COSMO G. MACKENZIE, JULIAN R. RACHELE (by invitation), NANCY CROSS (by invitation), JOSEPH P. CHANDLER, and VINCENT DU VIGNEAUD. *Department of Biochemistry, Cornell University Medical College, New York City.* The amount of CO_2 in the expired air derived from the oxidation of the methyl carbon of dietary methionine (labeled with C^{14} in the methyl group) was compared at two levels of methionine in the diet. Rats were allowed continuous access to a diet containing 0.4 per cent cystine, 0.2 per cent choline chloride, and either 0.6 or 1.2 per cent methionine. During the course of the experiment the animals were fed by stomzch tube a single 2 gm.-portion of the diet in which the ordinary methionine was replaced by radioactive methionine. On the low methionine diet 6%, and on the high methionine diet 25%, of the ingested methyl carbon appeared within 30 hours as radioactive CO_2 in the expired air. This difference in the proportion of the methyl group oxidized was due almost entirely to a difference in the relative rates of oxidation during the first 8 hours. On both diets, the rate of oxidation of the methyl group, as reflected in the $C^{14}O_2$ content of the expired air, fell into two phases. The first of these (period of assimilation) was characterized by a rapid initial rise in content to the third hour, followed by a decline until about the eighth hour. The second phase (period of equilibrium) was initiated at this time by the establishment of a relatively low and constant rate of oxidation that persisted for the remaining 22 hours of the experiment. The influence of other dietary constituents on the oxidation of the methyl group of methionine is being investigated.

Studies on the nitrogen metabolism of tomato using N^{15}-labeled ammonium sulfate. ROBERT MACVICAR (by invitation) and R. H. BURRIS. *Department of Biochemistry, University of Wisconsin.* Tomato plants were grown in sand culture with complete nutrient solution for six weeks; all nitrogen sources were then removed for ten days, at which time symptoms of nitrogen deprivation were appearing. 5.0 mg. of ammonium nitrogen as $(NH_4)_2SO_4$ containing 30 atom % N^{15} excess were supplied per plant at 7:00 A.M. Twelve hours later the plants were harvested, separated into aerial and root fractions, rapidly heated to 65° C., and dried. The dried tissue was then fractionated to obtain the principle nitrogenous fractions. The crude protein fraction was hydrolyzed and several amino acids and amino acid groups isolated from the hydrolysate. The N^{15} content of these fractions was determined by mass spectrographic analysis and the atom per cent N^{15} excess found to be:

Nitrogen fraction	Leaves and stem	Root
Total nitrogen	1.02	1.13
Ethanol-soluble	1.74	3.89
Water-soluble, ethanol-insoluble	0.49	0.77
Crude protein hydrolysate	0.58	0.79
Neuberg precipitate of hydrolysate	0.56	0.64
NH_3 and amide N of hydrolysate	0.72	0.38
Arginine	0.77	
Glutamic acid	4.16	1.15
Aspartic acid	0.94	0.75
Histidine	0.21	
Lysine	0.07	

The data indicate appreciable incorporation of the absorbed nitrogen into tissue protein during the 12-hour period and confirms the observations of Vickery, *et. al.* in tobacco (J. Biol. Chem., **135**: 531, 1940). The markedly higher concentration of labeled nitrogen in the dicarboxylic amino acids, and particularly in glutamic acid, indicates a high order of reactivity of these compounds with the administered ammonium nitrogen. Histidine and lysine are apparently synthesized less rapidly.

Effect of cytochrome C on the resistance of mice to anoxia. GEORGE H. MANGUN. *Department of Laboratories, Henry Ford Hospital, Detroit.* Experiments were designed to determine the effect of cytochrome *C* on the resistance of male CFW mice to anoxia and asphyxia. Animals injected intravenously via the tail vein with cytochrome *C* and an equal number of control animals injected with an equal volume of saline were placed together in pairs in a closed system of 500 cc. capacity 2 to 6 hours after injection, with or without absorption of CO_2, maintaining atmospheric pressure by the introduction of nitrogen. With dosages of 50–100 mg./kg. of cytochrome *C*, 6 controls and 6 cytochrome *C* injected animals exhibited symptoms and died simultaneously. At 200 mg./kg., results were somewhat variable, with 4 of the 6 cytochrome *C* injected animals dying 2–4 minutes earlier. At dosages of 300–450 mg./kg., 8 cytochrome *C* injected animals uniformly exhibited symptoms and succumbed before the controls. These experiments indicate that cytochrome *C* is unable to protect mice against the effects of a primary oxygen deficit. They are being repeated with other preparations of cytochrome *C*.

The chemical composition of human thoracic aortas. ROGER W. MARSTERS (by invitation), JACK R. LEONARDS (by invitation), and VICTOR C. MYERS. *Department of Clinical Biochemistry, Western Reserve University, Cleveland, Ohio.* Sixty-four human thoracic aortas have been analyzed for water, creatine, collagen, elastin, cholesterol, calcium, and phosphorus. The water content showed no significant change with age but it was decreased in severely arteriosclerotic specimens. Both creatine (as an index of smooth muscle) and elastin tended to decrease with age, while the collagen content increased; however, the differences are not statistically significant. The most outstanding changes were in the cholesterol, calcium and phosphorus contents of the aortas, marked increases in these three constituents occurring with age. There is a good correlation between the results of chemical analysis and the pathological diagnosis at autopsy. With increasing arteriosclerosis there are marked increases in cholesterol, calcium, and phosphorus content. In most cases these three constitutents increased in a parallel manner but occasionally the arteriosclerotic deposits were predominantly cholesterol.

Enzymatic destruction of cholesterol by rat liver extract in vitro. WALTER MARX and MORTIMER LIPSETT (introduced by HARRY J. DEUEL, JR.). *Department of Biochemistry, University of Southern California Medical School, Los Angeles.* It is known from balance experiments (Schoenheimer, Page and their co-workers) that the animal organism is able to dispose of cholesterol, when relatively large quantities are fed with the diet. The possibility was considered that this destruction of cholesterol is of an enzymatic nature, and it was attempted to demonstrate *in vitro* such an enzyme capable of destroying cholesterol. Various homogenates and extracts of rat liver and spleen were incubated with cholesterol under different conditions. Results of experiments using tissues from normal rats were equivocal. Liver extracts were then made from rats which had been fed a diet containing 1% cholesterol and 0.5% bile salt for periods of several weeks or months. The extracts were made using saline or phosphate buffer at pH 6 to 7, and they contained appreciable quantities of cholesterol. When such extracts were incubated at 37°C. for periods of 12 to 16 hours, a significant decrease in the concentration of total cholesterol.

The hydrolysis of nuclear proteins by cathepsins. I. Calf thymus cathepsin. MARY E. MAVER and ANTOINETTE GRECO (by invitation). *National Cancer Institute, Bethesda 14, Md.* The proteins of calf thymus, which were insoluble in 0.325MKCl and were sedimented at 2,000 r.p.m., were hydrolyzed by the cathepsins prepared from calf spleen and calf thymus with optima at pH 4.0 and at pH 6.0–6.5. The proteinase activity at pH 4.0 was activated by cysteine while that at the higher pH was not. This protein fraction yields both nucleic acid and histone abundantly. It contained approximately 15% nitrogen and 2 percent phosphorus. The nuclear protein or nucleo-histone fraction of calf thymus, separated by the usual method of solution in weak alkali and precipitation with acetic acid, was hydrolyzed at one pH optimum of

pH 4.5–5.0. The thymus histone and clupein, the protamine of herring sperm, were also hydrolyzed by calf spleen and thymus cathepsins.

Vitamin A deficiency and diet composition. JEAN MAYER (by invitation) and W. A. KREHL. *Yale Nutrition Laboratory, Department of Physiological Chemistry, Yale University.* Sprague Dawley weanlings of uniform weight were fed vitamin A free, synthetic diets; controls were kept for each group. Three diets were designed to study the influence of the protein level (10, 25 and 60% casein) on the growth and survival time of the deficient animals. Two high fat diets (purified lard and corn oil) were used to demonstrate the possible influence of saturated and unsaturated fats. Similarly dextrin was compared with sucrose. When casein replaces sucrose isocalorically, survival time varies inversely with the protein level and bears no direct relationship to the maximum weight attained. The difference in the average weights of the three control groups at 40 days was not statistically significant. Fats were found to be protective against vitamin A deficiency, as evidenced by an increased survival time. The average maximum growth of the deficient animals on hard fat was considerably and consistently better than on corn oil, although no significant growth difference was seen in the controls. The replacement of sucrose by dextrin did not result in any change. The authors have concomitantly demonstrated that vitamin C deficiency was an integral part of the vitamin A deficiency syndrome in the rat. The modification of the picture given above by addition of ascorbic acid is now being studied.

Pituitary-protein-bound iodine and the Plummer treatment of hyperthyroidism. J. F. MCCLENDON and WM. C. FOSTER (by invitation), Hahnemann Medical College, Phila. and J. W. CAVETT (by invitation). *Salisbury's Laboratories, Charles City, Iowa.* For several weeks 74 chickens were fed 0.2% thiouracil, 71 chickens received 4 cc. Lugol's solution per gallon drinking water and 71 were controls. The pituitaries of the controls contained an average of 0.052 micrograms of protein-bound iodine, PBI, (3 micrograms per gram fresh). The pituitaries of chickens fed thiouracil averaged 0.044 microgram PBI (2.5 micrograms per gram) and the pituitaries of chickens given Lugol's averaged 0.077 microgram PBI (3.8 micrograms per gram). The desiccated thyroids of the controls contained 0.199% PBI, those of the thiouracil-fed 0.0253% PBI and those of Lugol's-fed 0.372% PBI. This excess PBI in the thyroid apparently was not passed into the circulation as the chickens did not lose weight. Therefore we believe that the excess PBI in the pituitary has not migrated from the thyroid but is formed in situ (as McClendon and Foster postualted for sheep, Fed. Proc. **6**: 275) inside the pituitary, where it suppresses the formation of thyrotropin. Since the PBI cannot exceed the total iodine and the pituitary is more than 80% warer, our values are 5 times those of Closs who found 1 to 2 micrograms total iodine per gram dry pituitary. He spoke of "possibility of contamination" of commercial preparations 50 times his values. (It should be noted that U. S. P. thyroid contains 2000 micrograms per gram.) We interpret the variations of PBI in the pituitary as being formed during life and not due to contamination of the sample.

Methionine in the growth of the malarial parasite, *Plasmodium Knowlesi.* RALPH W. MCKEE and QUENTIN M. GEIMAN (by invitation). *Departments of Biological Chemistry and of Comparative Pathology and Tropical Medicine, Harvard Medical School, Boston, Massachusetts.* *Plasmodium knowlesi* requires the addition of methionine for its *in vitro* growth and multiplication (Fed. Proc., **6**, 276, 1947). This means that the parasite needs more methionine than it can obtain from the host red cell and from its immediate environment. Since this requirement is so critically important, it seemed desirable to determine, (1) possible changes in the amount of free methionine in the circulating plasma of the host animal, *Macaca mulatta*, and what effect this might have on the *in vivo* growth of the parasites and (2) the effects of methionine analogs and methyl-acceptor compounds on parasite growth and multiplication both *in vitro* and *in vivo*. The *P. knowlesi* infection in our monkeys is controlled by fasting the animal (not by subjecting the animal to lowered temperature, another stress condition) and is reversed by giving dl-methionine or para-aminobenzoic acid. This control of the parasitemia is not reversed by the oral administration of a body weight-maintenance level of sucrose or of sucrose plus injected ascorbic acid. Diets complete, except for methionine, are being tested. The plasma levels of methionine under these conditions are being determined. The *in vitro* growth of *P. knowlesi* is markedly inhibited by methoxinine and ethionine and the inhibition is overcome by the addition of extra methionine. The antagonist/methionine ratios are about 10/1 and 1/1, respectively. Guanidoacetic acid, an acceptor of methyl groups from methionine is being tested in monkeys, both for its effect on parasite growth and on the plasma level of methionine.

A crystalline compound of β-Lactoglobulin with dodecyl sulfate. T. L. MCMEEKIN, B. D. POLIS (by invitation), E. S. DELLAMONICA (by invitation) and J. H. CUSTER (by invitation). *Eastern Regional Research Laboratory, Philadelphia 18, Pa.* A crystalline derivative of β-lactoglobulin containing two equivalents of dodecyl sulfate has been prepared. The compound is apparently undissociated, since the dodecyl sulfate is not removed

by barium ion. The iso-electric point of the dodecyl derivative is slightly more acid than that of β-lactoglobulin, as shown by its mobility. Moreover, a salt solution of the dodecyl derivative of β-lactoglobulin has a pH of 5.06, as compared with 5.18 for β-lactoglobulin. The solubility of the dodecyl derivative is about one-half as large in water and one-third as large in dilute salt solutions as that of β-lactoglobulin. The titration curves of both these substances are essentially the same on the acid side of the iso-electric point, while on the alkaline side the titration curve of the dodecyl derivative indicates a greater alkali-combining capacity. This finding is in agreement with the increased mobility obtained with the dodecyl derivative, as compared with that of β-lactoglobulin on the alkaline side of the iso-electric point. The dodecyl derivative has the same amount of amino nitrogen as β-lactoglobulin (on a total N basis), as indicated by the Van Slyke method. This dodecyl derivative did not give a test for sulfhydryl groups. Efforts to remove the two equivalents of dodecyl sulfate from β-lactoglobulin have failed, indicateing that previous reports of the complete removal of this substance from proteins are in error, since the mobilities of the proteins after treatment with dodecyl sulfate and subsequent removal are invariably different from those of the untreated proteins.

An egg yolk protein containing 10% phosphorus. DALE K. MECHAM (by invitation) and HAROLD S. OLCOTT. *Western Regional Research Laboratory, Albany, Calif. Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Department of Agriculture.* A protein preparation containing 10% phosphorus is obtainable from egg yolk as follows: Diluted yolk is centrifuged (Sharples). The insoluble fraction is lyophilized, extracted with ether, and dispersed in 5% sodium sulfate. Addition of sodium sulfate to a concentration of 36% precipitates the bulk of the proteins. The phosphoprotein is precipitated from the remaining solution with copper acetate. The precipitate is dissolved at pH 2 and reprecipitated at neutrality. After further purification steps, the copper is removed by dialysis against citrate buffer (pH 5). One preparation appeared to be 90% homogeneous by electrophoretic analysis. The following properties are typical: nitrogen, 12%; phosphorus, 10%; sulfur, none; number average molecular weight (osmotic pressure), 18,000 to 25,000. Titration data indicate that the phosphorus occurs as ortho-phosphate ester. Sufficient serine is present to account for at least 80% of the phosphorus, and the presence of *o*-phosphate esters of serine residues is further suggested by a marked destruction of serine during acid hydrolysis, and by the development of increased ultra-violet absorption, characteristic of dehydroalanine residues, after alkaline dephosphorylation. The phosphoprotein resists digestion by whole pancreas extract but becomes digestible after removal of the phosphate groups with phosphatase (orange or grapefruit). At least 40%, and possibly considerably more, of the total protein phosphorus content of egg yolk can be accounted for in the new phosphoprotein, for which the name "phosvitin" is proposed.

Enzymatic hydrolysis of acetopyruvic acid. ALTON MEISTER (by invitation) and JESSE P. GREENSTEIN. *National Cancer Institute, National Institute of Health, Bethesda, Maryland.* Acetopyruvic acid (α,γ-diketovalerianic acid) is rapidly hydrolyzed to yield nearly equivalent amounts of pyruvic acid equally well in aerobic or anaerobic digests with aqueous extracts of rat liver and kidney. The optimum pH for the reaction is about 7.0. Prolonged dialysis of the extracts does not appreciably affect the reaction. There is no detectable acetoacetate formation. The resulting pyruvic acid is measured as the 2:4 dinitrophenylhydrazone and has been identified by isolation. Rat spleen, pancreas, brain, skeletal and cardiac muscle, testes, intestine, stomach, lung, and primary hepatoma show negligible activity. Acetopyruvate accelerates the desamidation of glutamine by rat liver to nearly the same extent as does the equivalent amount of pyruvate. However, no acceleration of glutamine desamidation by acetopyruvate was noted in the rat hepatoma, thus indicating that the diketo acid itself is not active in the glutamine system. Freshly prepared acetopyruvic acid was employed as the purified sodium salt. This salt, the free acid obtained therefrom, and the ethyl ester of the acid all possess a characterisitc absorption in the ultraviolet with a maximum at 2950Å for the Na salt, 2850A for the free acid, and 2900Å for the ester.

Inhibition of hyaluronidase by hydroquinones and quinones. KARL MEYER and CHARLES RAGAN (by invitation) with the technical assistance of HANNAH WEINSHELBAUM. *Departments of Ophthalmology and Medicine, School of Medicine, Columbia University, New York.* The inhibition by salicylate of the spreading reaction in animals and in man (Guerra, F. J. Pharm. Exp. Therap. **87**: 1943, 1946) has been confirmed. However, in vitro salicylate in therapeutic levels had no effect on hyaluronidase activity (Meyer, K. Physiol. Revs. **27**: 335, 1947; Pike, R. M. Science. **105**: 391, 1947; Dorfman, A. et al. Proc. Soc. Exp. Biol. and Med. **64**: 357, 1947) but did inhibit in concentrations which denatured the protein. The urine of patients on salicylate therapy was fractionated and the fractions tested for inhibition of hyaluronidase. A potent inhibitor was found in the recrystallized salicyluric acid fractions, but synthetic salicyluric acid had no effect. The inhibition proved to be due to gentisic and gentisuric acids. Synthetic gentisic

acid after short incubation with hyaluronidase gave strong inhibtion which could be prevented by hyaluronate. This observation is in agreement with the starement that 2-5-benzoquinone carboxylic acid inhibits hyaluronidsae (Lowenthal, J. and Gagnon, A. Science. **105:** 618, 1947). The following hydroquinones and quinones strongly inhibited hyaluronidase in 0.0005 mol concentration: hydroquinone dicarboxylic acid (25%), synthetic gentisic acid (44%), homogentisic acid (59%) and polyporic acid (93% inhibition). The significance of the findings will be discussed.

The free energy of phosphorylation. Otto Meyerhof and Peter Oesper (by invitation). *Department of Physiological Chemistry, School of Medicine, University of Pennsylvania.* From equilibrium measurements by means of phosphatase (Schmidt's intestinal phosphatase) $\Delta F°$ for hydrolysis of primary alcoholic esterphosphate in hexosephosphates, D-3-phosphoglyceric acid, etc. is found to be -300 to -500 cals. By means of the equilibrium of the reaction: phosphopyruvate + ADP $\rightleftharpoons$ pyruvate +ATP, $\Delta F°$ (from left to right) is found to be about -3000 to -3500 cals. The same $\Delta F°$ of transphosphorylation obtains from 1,3-diphosphoglyceric acid to ATP. From these and other equilibria, which were measured formerly by the authors and others, four classes of phosphate compounds can be distinguished with regard to the $\Delta F°$ (25°C.; pH 8) of phosphorylation; they are in decreasing order:

1. Acylphosphates and enolphosphates, $\Delta F°$ around -14000 cals.
2. Pyrophosphates and guanidinophosphates, $\Delta F°$ around -10500 cals.
3. Carbonylphosphate (Glucose-1-phosphate), $\Delta F°$ around -2000 cals.
4. Primary alcoholic phosphates, $\Delta F°$ around -400 cals.

The bearing of these data for the energy transfer will be discussed.

Loss and restoration of rat liver enzymes (activity) related to dietary changes in liver protein. Leon L. Miller. *Department of Biochemistry, The Jefferson Medical College, Philadelphia, Pa.* The activity of five enzymes (catalase, alkaline phosphatase, xanthine dehydrogenase, cathepsin, and arginase) has been measured by using homogenates of livers from Sherman strain rats. When compared with the livers of normal controls on a 25% casein diet adequate in vitamins, the livers of animals on a non-protein diet for 8 to 18 days show a loss of activity of all the enzymes studied. The loss of activity is greatest when expressed in terms of units of activity per 100 grams of initial body weight, but is highly significant when expressed in units per gram of liver protein (N × 6.25). The loss of enzyme activity is most probably the result of the loss of enzyme protein per se. On this basis presumably vital enzyme proteins are not spared, but are lost along with liver cell protein in general. A small group of rats was refed a 25% casein diet after 30 days on a 6% casein diet followed by 5 to 9 days on the non-protein diet. In most of the rats there was prompt restoration toward normal of liver protein and enzyme protein (i.e. activity).

Utilization of pyruvate by heart ventricle *in vitro* as a function of time. O. N. Miller (by invitation), R. E. Olson (by invitation) and F. J. Stare. *Department of Nutrition, Harvard School of Public Health and Department of Biological Chemistry, Harvard Medical School, Boston.* The rate of pyruvate utilization by cells *in vitro* has been determined for a variety of animal tissues, eggs, and microorganisms by a number of investigators. The object of the present study was to determine the effect of the length of the period of measurement and the time of substrate addition upon the rate of pyruvate disappearance and lactate production in slices of ventricle from cardiac muscle of well nourished rats and ducks. Slices of ventricle were bathed in phosphate-saline containing 5 mM/L of pyruvate for varying intervals in a standard Warburg apparatus at 37° in an atmosphere of oxygen. In some experiments pyruvate was initially present; in others it was added after different intervals. Pyruvate and lactate were determined chemically and the rates of pyruvate disappearance and lactate production expressed as (Q). The following table gives the results obtained.

Time of measurement	$-Q_{pyruvate}$	$+Q_{lactate}$
minutes		
5–15	>20	>10
15–30	14–20	5–10
30–60	8–14	3– 5
60–120	5– 8	2– 3
120–240	<5	<2

The values obtained with rat and duck ventricle are included together in the above ranges although, in general, the rat values were higher than the duck values. Comparable decreases in pyruvate utilization were obtained by adding pyruvate after given intervals of incubation without added substrate.

Chromatography of amino acids. Colorimetric ninhydrin method for analysis of the effluent. Stanford Moore (by invitation) and William H. Stein. *Rockefeller Institute for Medical Research, New York City.* The reaction of ninhydrin with NH_2-groups to give diketohydrindylidene-diketohydrindamine has been utilized as the basis for a colorimetric determination of amino acids and related compounds in effluent samples from strach chromatograms. The lack of reproducibility of the color yield from a given amino acid in the ninhydrin reaction, and the marked deviations from Beer's law which have been observed, are overcome by the

incorporation of hydrindantin or stannous chloride in the reagent solution to eliminate oxidative side reactions. Although under these conditions the color yield from a given amino acid is constant, the different amino acids do not all give the same percentage yield of the blue product. This fact does not prevent the accurate use of the method in chromatographic work in those cases where the individual amino acids are separated from one another by the fractionation process. The ninhydrin reaction is carried out at pH 5 and 100°C. The absorption maximum of the colored product is at 570mμ for all the α-NH_2 acids (except cysteine), whereas with the β-napthoquinone sulfonic acid reaction, the maxima are similar but not identical. On individual amino acids the accuracy is ±2 per cent with samples containing 3 micrograms of α-NH_2 nitrogen.

The color development is obtained with a variety of compounds containing NH_2-groups, including amino acids, peptides, primary amines, and ammonia. For chromatographic work the generality of the reaction extends its usefulness. For work with unfractionated biological material the lack of specificity is a disadvantage.

The purification and crystallization of phosphoglucomutase. VICTOR A. NAJJAR. *Department of Biological Chemistry, Washington University Medical School.* Perfused rabbit muscle is passed through a meat grinder and extracted with two volumes of water (I), adjusted to pH 5 with molar acetic acid, heated to 65°C. and filtered. The filtrate (II) is made to 0.65 ammonium sulfate saturation. The precipitate is dissolved in acetate buffer pH 5.0 (III) then heated to 63°C. and centrifuged. The supernatant (IV) is raised to an ammonium sulfate concentration of 0.50 and precipitate discarded. The concentration is raised gradually to 0.55 where crystals appear after a few hours. The saturation is further raised to 0.60 and allowed to stand 24 hours. The 0.50–0.60 crystals (V) are then separated, dissolved in acetate buffer and recrystallized with ammonium sulfate. The crystalline fraction between 0.55–0.60 (VI) is approximately 90% pure and that between 0.60–0.65 (VII) is electrophoretically homogenous at pH 5.0.

	Fractions						
	I	II	III	IV	V	VI	VII
Activity in units/mg. protein	0.5	3.0	6.0	14.0	21	23	26
Phosphoglucomutase protein (mg.)	400	200	180	140	130	19	20

The enzymatic hydrolysis of glutathione. KAY NAKAMURA (by invitation) and FRANCIS BINKLEY. *Laboratory for the Study of Hereditary and Metabolic Disorders and the Departments of Biochemistry and Medicine, University of Utah School of Medicine, Salt Lake City, Utah.* It has been found that glutathione is hydrolyzed to cysteinylglycine and glutamic acid by an enzyme of kidney tissue. This enzyme is relatively stable to heat and is activated by magnesium ions. The enzyme responsible for the hydrolysis of cysteinylglycine to cysteine is found in liver and muscle as well as in the kidney. This enzyme is labile to heat and is strongly inhibited by calcium or magnesium ions. Desiccated hog kidney (Viobin Corporation) is a good source of the enzyme responsible for the hydrolysis of glutathione to cysteinylglycine; considerable purification of this enzyme has been achieved. The purified enzyme has been utilized in the estimation of glutathione in whole blood and in homogenates of tissue. The cyxteinylglycine is measured by the method of Sullivan and Hess (J. Biol. Chem., **116**, 221 (1936)) for cysteine. Glutathione, added to whole blood or homogenates of tissues, may be quantitatively recovered by this method.

Blood sugar changes following the administration of lactose in raw and evaporated milk. SAMUEL NATELSON (by invitation), BENJAMIN KRAMER and MARVIN SHERMAN (by invitation). *Pediatric Research Laboratory, Jewish Hospital of Brooklyn.* The same infants on successive days (3rd and 4th day after birth) show fairly close correlation in carbohydrate tolerance curves when the same carbohydrate is administered as measured by the concentration of reducing sugar in blood. The following formulae were compared in the same infant by measuring carbohydrate tolerance curves on successive days: a). Lactose in evaporated milk formula vs. Lactose in raw cow's milk formula; b). Starch hydrolysate (Cartose) in evaporated milk formula vs. Starch hydrolysate in raw cow's milk formula; c). Raw mother's milk. Lactose in raw milk formula shows higher peaks and more rapid return to the fasting level than Lactose in evaporated milk formula. Starch hydrolysate in evaporated milk formula shows higher peaks and a more rapid return to the fasting level than lactose in the same formula, but improvement is observed when the starch hydrolysate is administered in raw milk. Raw mother's milk per se shows higher peaks than any of the above formulae. The time required for return to fasting level was the same as for lactose in raw cow's milk. These observations may explain our statistical growth studies which show improved weight curves for infants, in the first seven days of life, on breast milk supplemented with starch hydrolysate (Cartose) over evaporated milk supplemented with starch hydrolysate.

Response of citric acid levels to administration of glucose. SAMUEL NATELSON, JOSEPH B. PINCUS and JULIUS K. LUGOVOY (introduced by ALBERT

E. SOBEL). *Department of Biochemistry of the Jewish Hospital of Brooklyn.* Following the administration of glucose orally the citric acid level, in serum, drops slowly for the first thirty minutes and then drops sharply to a minimum point (25–35% lower than the fasting level) about 140–200 minutes after glucose administration. A gradual return, after 5–6 hours, to the fasting level is then observed. The minimum for citric acid corresponds with the period of most rapid drop in glucose level. Injection of insulin, without glucose administration, causes a drop in citric acid serum level. Most diabetics exhibit a citric acid response which resembles the normal. Certain convulsive children and diabetics show an abnormal citric acid response. This abnormality takes the form of a rising level of citric acid which does not fall during the test period. Another form of abnormality was a sharp rise for the first hour followed by a lowering of the citric acid level to a minimum below the fasting level. An *in vivo* relationship between the intermediate metabolism of glucose and citric acid is therefore indicated, which may be disturbed in certain pathological conditions.

The enzymatic liberation of pantothenic acid. J. B. NEILANDS (by invitation) and F. M. STRONG. *Department of Biochemistry, University of Wisconsin, Madison, Wisconsin.* The purified coenzyme for acetylation (J. Biol. Chem. **162,** 743, 1946) was found by Lipmann and his associates (J. Biol. Chem. **167,** 869, 1947) to contain pantothenic acid. In this form the pantothenate is unavailable to *L. casei* although it may be liberated by the simultaneous action of a purified alkaline intestinal phosphatase and a pigeon-liver enzyme. The use of these enzyme treatments in the preparation of samples for pantothenic acid assay has been compared with the mylase P digestion method (Arch. Biochem. **9,** 251, 1946). In general the new procedure gives approximately a 3-fold increase in the apparent pantothenic acid level, although thirty-five fold increases have been obtained by allowing the enzymes to act on a boiled extract of chicken liver. The latter was prepared immediately after death. The pigeon-liver enzyme has been found to be replaceable by a chicken-liver enzyme; the latter is a more convenient source for routine assay work. It would appear that a large share of the pantothenic acid present in a wide variety of foods and other biological materials exists in the combined form, and has not been included in most of the previously reported assay values.

Kinetics and inhibition of carboxypeptidase activity. HANS NEURATH and ELAINE ELKINS (by invitation). *Department of Biochemistry, Duke University School of Medicine, Durham, N. C.* Independent determinations of the proteolytic coefficient ($C = k/E$) for the hydrolysis of carbobenzoxyglycylphenylalanine (CGP) by carboxypeptidase, in 0.05 M substrate solutions, have yielded a value of about $C = 10$. Present measurements reveal a large dependence of the first order reaction constant, k, on substrate concentration (DL-CGP). The corresponding proteolytic coefficients approach a minimum value of $C = 5$ above 0.075 M substrate (with respect to L-CGP) and a maximum value of $C = 53$ below 0.001 M substrate. A similar trend was observed with solutions of L-CGP. The data fit closely the equations of Lineweaver and Burk. None of the reaction products, i.e. carbobenzoxyglycine and L-phenylalanine, inhibits hydrolysis nor does D-CGP. However, hydrolysis is largely inhibited by D-phenylalanine. The possibility of optical inversion during enzymatic hydrolysis has been excluded by the observation that phenylalanine isolated from the enzymatic hydrolysate of L-CGP is not oxidized by D-amino acid oxidase. It appears, therefore, that the dependence of reaction rates on substrate concentration is unrelated to the inhibition by D-phenylalanine. This conclusion is further corroborated by the findings that the latter inhibition is of the non-competitive type. While D-lysine is not inhibitory, the extent of inhibition by other D-amino acids decreases in the order: phenylalanine > histidine > valine = isoleucine. It is noteworthy that these D-isomers are inhibitory despite the fact that peptides containing some of the corresponding L-isomers are not specific substrates for carboxypeptidase.

Estrogenic activity of doisynolic acid and related compounds. HAROLD J. NICHOLAS (by invitation), SIDNEY A. THAYER and EDWARD A. DOISY. *Laboratory of Biological Chemistry, Saint Louis University School of Medicine, Saint Louis, Missouri.* Since Miescher's discovery of the pronounced estrogenic activity of 1-ethyl-2-methyl-7-hydroxy - 1,2,3,4,9,10,11,12 - octahydrophenanthryl-2-carboxylic acid (doisynolic acid) and some related compounds, we have reinvestigated some of these derivatives in our laboratory. Doisynolic acid and the 7-benzoate methyl ester of this compound have been assayed in spayed rats and mice. Similarly bisdehydrodoisynolic acid and its methyl ether have been assayed. We find that doisynolic acid injected subcutaneously is less active than estrone in spayed rats. However, this acid is more active than estrone administered orally to spayed rats. It is considerably less active than estrone (approximately 1/150 as active) by subcutaneous injection in spayed mice, an observation which agrees with out earlier assay of this substance. Doisynolic acid possesses about the same order of activity as estrone when given orally to mice. These data again emphasize the difficulty in attempting to compare the activity of one compound with another estrogenic material using only one species of animal. From the mixture resulting from

fusion of estrone with potassium hydroxide we have isolated doisynolic acid, estrone and another steroid insoluble in carbonate.

Respiratory metabolism in yeast and coenzyme A levels. Effect of phenyl panthenone on coenzyme A synthesis. G. DAVID NOVELLI (by invitation) and FRITZ LIPMANN. *Biochemical Research Laboratory, Massachusetts General Hospital, and the Department of Biological Chemistry, Harvard Medical School, Boston.* Samples of yeast, high and low in coenzyme A (Co A), were obtained from pantothenic acid deficient yeast by pre-treatment, in a glucose-phosphate medium, with and without pantothenic acid. In this manner samples could be compared containing an average of 350 and 100 units of Co A respectively. The respiration of acetate, ethanol and glucose was followed; increase in Co A corresponded with a doubling to tripling of the respiratory rate with acetate and ethanol and about 30% increase with glucose. In the deficient samples an accumulation of acetic acid occurred with ethanol and glucose. Malonate depressed the stimulated respiration of acetate while there was little effect in the deficient sample. Preliminary results have been obtained in a study with dried yeast of citrate synthesis from acetate and oxalacetate. The described yeast system offers an opportunity to study the mode of action of pantothenic acid-antimetabolites. Mainly phenyl panthenone (Wooley et. al. J. Biol. Chem., **159**: 263, 1945) was studied. Its presence, together with pantothenic acid, in the pre-treatment period, was found to suppress completely Co A synthesis yielding a preparation with low acetate oxidation. When added, however, after the enrichment period, acetate oxidation was unaffected or rather stimulated by addition of phenylpanthenone. The observation indicates the action of phenylpanthenone to be due to a blocking of the synthesis of Coenzyme A from pantothenic acid.

Effect of biotin-deficiency upon the respiration of cardiac muscle in ducklings. R. E. OLSON (by invitation), O. N. MILLER (by invitation), and F. J. STARE. *Department of Nutrition, Harvard School of Public Health and Department of Biological Chemistry, Harvard Medical School, Boston.* A number of recent reports have indicated that biotin functions in metabolism to facilitate the reversible carboxylation of pyruvic acid to oxalacetic acid. If the failure to accomplish this reaction were a major biochemical lesion in biotin deficient tissues it might be expected that the rate of utilization of pyruvate as well as intermediates of the tricarboxylic acid cycle would be depressed. To test this hypothesis we have determined the rate of oxygen consumption of slices of ventricle from hearts of biotin-deficient and normal ducks without added substrate and with added pyruvate and succinate. The rate of disappearance of pyruvate during the period of respiration was determined chemically and the rate of succinate metabolism was measured by determining the rate of radioactive CO_2 production from carboxyl labeled succinate during the period of respiration. The oxygen consumption of biotin deficient heart ventricle is markedly depressed. Without added substrate it is reduced 30%, with added pyruvate 27% and with added succinate 23%. The rate of disappearance of pyruvate to non-lactate products is reduced 65% and the rate of CO_2 production from succinate is reduced 45%. The injection of biotin to deficient birds restores these values approximately to normal.

Further studies on the effect of cysteine and histidine on the production of cobalt polycythemia. JAMES M. ORTEN. *From the Department of Physiological Chemistry, Wayne University College of Medicine, Detroit.* Earlier studies in this laboratory have demonstrated that the oral administration to rats of cysteine or of histidine significantly inhibit the production of polycythemia by cobalt also given orally. On the other hand, methionine and choline in equivalent amounts had no detectable effect. Since both cysteine and histidine form less soluble complexes with cobalt than inorganic cobalt salts, the question was raised as to whether the inhibiting effect of these two amino acids might be explained on the basis of a decrease in the intestinal absorption of cobalt. In order to study this question further, the complexes of cobalt with cysteine and with histidine have been prepared and injected subcutaneously into rats in amounts equivalent to 1.0 mg. cobalt per day. Control rats were injected the same amount of cobalt as the sulfate. A 12-week period of observation was employed during which time the hemoglobin content of the blood was determined biweekly. The data obtained thus far indicate that cobalt injected as the cysteine complex does not produce a polycythemia. Studies with the histidine complex are in progress. The relation of these observations to the possible mechanism of cobalt action in increasing hemopoiesis will be discussed.

Peptidase increase during growth of the salivary glands of the fly, Drosophila melanogaster. ELIZABETH K. PATTERSON, MARJORIE E. DACKERMAN, and JACK SCHULTZ (introduced by THEODORE F. LAVINE). *Lankenau Hospital Research Institute and Institute for Cancer Research, Philadelphia, Pa.* Peptide splitting enzymes have been studied during growth of the salivary glands of various genetic stocks of Drosophila melanogaster. Using Linderstrøm-Lang-Holter micro-techniques extracts of the glands are found to have the ability to split alanylglycine, leucylglycine, leucylglyclyglycine, and triglycine in order respective to their activity. Action of activators and inhibitors, pH activity, concentration, and time curves show the

enzyme splitting alanylglycine to be a typical intracellular peptidase. Alanylglycine is used as the substrate in the subsequent experiments on single glands. The larval salivary glands reach their maximum size in the early prepupa and then disintegrate. Cell division stops in the embryo and therefore growth of a constant number of cells of like physiological function may be studied. Comparison of different stocks attaining diverse nitrogen levels at maximum growth shows the peptidase content of the glands to be proportional to their nitrogen content. During growth the ability of the glands to split alanylglycine increases with their nitrogen content. As the glands break down, a sharp decrease in splitting ability occurs. The results show a correlation of peptidase content with growth of the cells and not with their breakdown.

The isolation of pregnanol-3(α)-one-20, Comp'd Y ($C_{21}H_{36}O_2$) and Comp'd Z ($C_{19}H_{32}O_2$) from the bile of pregnant cows. W. H. PEARLMAN. *Department of Biochemistry, Jefferson Medical College, Philadelphia.* That batch (31.0 liters) of bile from which estrone had previously been isolated (Pearlman et al., J. Biol. Chem., **170:** 173 (1947)) was further investigated. The ether-soluble neutral material of unhydrolyzed bile was repeatedly partitioned between petroleum ether and 90% methanol. The latter fraction was separated into ketonic and non-ketonic moieties with the aid of Girard's reagent T. The ketones were treated with succinic anhydride to obtain the alcoholic ketonic fraction which, following the removal of a small quantity of digitonin-precipitable material, was chromatographed. There were obtained by these procedures 15 mg. of pregnanol-3(α)-one-20. It was identified by its melting-point, specific optical rotation and C,H analysis; a diacetyl derivative was also prepared and similarly identified. The non-ketonic fraction, on treatment with ethyl acetate, yielded 142 mg. of a white crystalline product, m.p. 200–208° which proved to be a mixture and was therefore chromatographed. Comp'd Z, m.p. 229–229.5° $[\alpha]_D = -9°$ (dioxan) was thereby obtained., Found: 78.15% C, 11.07% H; $C_{19}H_{32}O_2$, calculated, 78.03% C, 11.03% H. Comp'd Y, m.p. 234–236°, $[\alpha]_D + 19°$ (dioxan) was also obtained. An additional 33.0 liters of bile was obtained from pregnant cows and worked up as before. There were obtained 20 mg. of Comp'd Z, m.p. 228.5–229°, $[\alpha]_D -2°$ (ethanol); found: 78.12% C, 11.05% H. There were also obtained 16 mg. of Comp'd Y (rectangular plates from alcohol), m.p. 235–236°, $[\alpha]_D +20°$ (abs. ethanol). Found: 78.73% C, 11.25% H; $C_{21}H_{36}O_2$, calculated, 78.69%, 11.33% H. Comp'ds Y and Z are non-digitonin precipitable. Comp'd Y is not identical with pregnanediol-3(α),20(α). Structural elucidation of these products is in progress.

Preliminary observations on the renal excretion of radioiodine after administration of tracer doses. MARSCHELLE H. POWER, WILLIAM C. MCCONAHEY, JR. (by invitation), F. RAYMOND KEATING, JR. (by invitation) and JOSEPH BERKSON (by invitation). *Divisions of Biochemistry, of Medicine and of Biometry and Medical Statistics, Mayo Foundation and Mayo Clinic, Rochester, Minnesota.* Tracer doses equivalent to about 100 micro-curies of radioactive iodide (I^{131}, Oak Ridge Operations, Atomic Energy Commission), together with 100 micrograms of nonradioactive sodium iodide, have been ingested by human beings. Subsequently samples of blood plasma and of urine, collected at suitable intervals, have been analyzed for their content of I^{131}. Plasma clearances of I^{131}, calculated from these data by the conventional procedure, that is, by multiplying the ratio of the concentrations of I^{131} in urine and plasma by volume of urine per minute range from about 40 cc. per minute in normal persons down to 15 cc. or less in persons with varying degrees of renal insufficiency. An alternate method of calculation involves extrapolation to zero time of the plot of log plasma I^{131} concentrations versus time, and division of the (hypothetical) value for plasma I^{131} so obtained into a quantity representing rate of renal excretion of I^{131}. (The latter value is derived by the procedure of Keating, Power, Berkson and Haines, Journal of Clinical Investigation, in press.) The values for plasma clearance of I^{131} obtained by this method are essentially similar to those obtained by the conventional method. These preliminary observations indicate, insofar as the clearance of tracer amounts of I^{131} may be accepted as a valid measure of the clearance of the nonradioactive inorganic iodide of the plasma, that the clearance of the latter under the conditions of our observations is considerably greater than that of the vastly more abundant halogen, chloride.

N-acylated and N-methylated glycyldehydroalanine and related compounds. VINCENT E. PRICE (by invitation) and JESSE P. GREENSTEIN. *National Cancer Institute, National Institute of Health, Bethesda, Maryland.* Chloroacetylglycyldehydroalanine, glycylglycyldehydroalanine, and N-methylglycyldehydroalanine, in which one hydrogen atom of the α-amino group is substituted, and chloroacetyl - N - methylglycyldehydroalanine in which both hydrogen atoms of the amino group are substituted, were prepared and subjected to exzymatic hydrolysis at pH 8 in certain rat tissue digests. The first two compounds were hydrolyzed in all tissue digests at close to the same rate as glycyldehydroalanine and were presumbaly attacked by dehydropeptidase I. No significant increase in α-amino nitrogen was noted in ammonia-free, kidney digests of chloroacetylglycyldehydroalanine after the dehydropeptide bond was nearly completely hydrolyzed. N-methylglycyldehydro-

alanine was hydrolyzed at a lower rate than glycyldehydroalanine, while chloroacetyl - N - methylglycyldehydroalanine was not hydrolyzed in any of the tissues studied. To serve therefore as a substrate for dehydropeptidase I, the compound need not possess a free, basic α-amino group, but it must evidently have an α-nitrogen atom to which at least one hydrogen is attached. Under certain conditions, reaction of chloroacetyldehydroalanine with methylamine or dimethylamine leads not only to replacement of the halogen but also to substitution at the double bond.

Purification and properties of *E. coli* bacteriophage T_6. FRANK W. PUTNAM, LLOYD M. KOZLOFF (by invitation) and E. A. EVANS, JR. *Department of Biochemistry, University of Chicago.* Study of protein synthesis and nucleoprotein turnover in virus-infected organisms has been initiated using the *E. coli* bacteriophage system as a model. By differential centrifugation in the Sharples supercentrifuge and in the high-speed angle centrifuge, bacteriophage T_6r^+ parasitizing *E. coli* (Strain B) has been isolated in a yield exceeding 8 mg. per liter of crude lysate. For phage purified from nutrient broth medium the infectivity (gm. N per infectious unit) measured by the plaque count method averaged $10^{-15.84}$. Elemental and component analysis, including P partition of the purified phage has been made. Desoxyribonucleic acid (DNA) is the characteristic nucleic acid. Physical characterization of the purified phage is being undertaken. Factors stimulating or inhibiting virus reproduction in broth or in synthetic (lactate) medium have been studied by the one-step growth curve technique. While common metabolic inhibitors such as cyanide, azide, *etc.* only partially repress growth or lysis, addition of a cationic detergent (1:10,000) toward the end of the latent period completely inhibits phage reproduction. Phage containing radioactive phosphorus (infectivity $10^{-15.75}$, activity 66 counts/microgram P) has been isolated from a broth lysate containing 0.46 microcuries P^{32}/ml., and the specific activity of the P fractions measured. Preliminary experiments indicate that the specific activity of the DNA fractions of the phage and the host bacterium are similar. Further radioisotope studies of the mechanism of phage synthesis are in progress.

Analytical procedure for determining individual tocopherols in a mixture of α-, β-, γ-, and δ-tocopherols. M. L. QUAIFE (introduced by P. L. HARRIS). *Research Laboratories of Distillation Products, Inc. Rochester, New York.* A method for determining β-, γ-, and δ-tocopherols in a mixture containing all three as well as α-tocopherol is presented. It depends on the formation of their yellow nitroso derivatives. These are separated by a simple chromatographic step and measured photometrically. The nitrosotocopherols are formed by treating β-, γ-, and δ-tocopherols in alcoholic solution with nitrous acid. Alpha-tocopherol gives no colored product. They are red in alkaline solution but revert to yellow when extracted into petroleum ether. They are measured with a Beckman spectrophotometer at 410 mμ or with an Evelyn photometer, using filter 400. Under the conditions used γ- and δ-nitrosotocopherols have the same extinction, while β-nitrosotocopherol has about one-third less. The nitrosotocopherols can be separated by a simple chromatographic step and evaluated. They form separate red bands on the column with the γ-derivative at the bottom, the β- in the middle, and the δ- (two bands) at the top. These are eluted separately according to the flowing chromatogram technique. Residues of the eluates are dissolved in petroleum ether and their absorption measured as noted. Spectra of the eluates were found to be identical to those of control unchromatogrammed solutions of the nitrosotocopherols. Synthetic mixtures of all three non-α-tocopherols in the presence of α-tocopherol have been separated and the components recovered quantitatively. The method has been applied to commercial vitamin E concentrates which contain all four tocopherols and to vegetable oils, following laboratory molecular distillation.

Gas pressure regulation in flame photometry. JOS. M. QUASHNOCK (introduced by WM. S. MCELLROY). *Department of Physiological Chemistry, School of Medicine, University of Pittsburgh.* A Perkin-Elmer flame photometer with a Rangelator 106-64 was studied to measure the degree of gas pressure regulation and the influence of any variation on the accuracy of the determinations. Gas pressure was measured for four hour periods while the photometer was in use. Maximum pressure change was 0.8 mm. water with a mean working pressure of 81.0 mm. The mean time elapsing between successive minimum and maximum pressures was 5.4 seconds. Measurements were then made on Na and K standards corresponding to normal sera diluted 1:500 for Na and 3:100 for K. The instrument was set at 100 with 10 p.p.m. Na or K standard. The various concentrations were determined at gas pressures of 80 mm. and 78 mm. water in rapid succession. This pressure change gave greatest differences in measurements of high concentrations. At K concentration corresponding to 8.5 meq./l. the results were 4.2% lower for the 78 mm. pressure than for 80 mm. Na measurements at 217.5 meq./l. were 2% apart. Variations in measurements of Na in the normal range of concentrations were less than 1%, while the differences for K were too small to be measureable. Since the maximum pressure change under operating conditions is 0.8 mm. water, the actual error will be less than half of the above. The gas pressure regulator studied is adequate and gives variations which do

not significantly influence the accuracy of the flame photometer for measurements of normal serum Na and K.

Role of platelets in coagulation of blood evidence of an inhibitor of the platelet factor. ARMAND J. QUICK. *Department of Biochemistry, Marquette University School of Medicine, Milwaukee.* It has recently been found that platelets do not supply thromboplastin, but an agent which activates the thromboplastin precursor (thromboplastinogen). When blood is collected under conditions in which the platelets are scrupulously preserved (i.e. at low temperature and with methylchlorosilane coated instruments and containers) and is centrifuged at high speed to remove the platelets, a plasma is obtained which clots very slowly and in which little prothrombin is converted to thrombin. When washed hemophilic platelets are added to such a plasma, a normal coagulation time and a significant increase in prothrombin consumption results. Likewise, the addition of this platelet-free plasma to hemophilic blood normalizes the coagulation time and increases the prothrombin consumption. This is accomplished by supplying thromboplastinogen which hemophilic blood lacks. There is no evidence that it contains any inhibitory agent. Blood obtained from a patient 69 years old, who developed a hemophilia-like disease following a dermatitis, had a coagulation time of 35 minutes. When this blood was added to an equal volume of normal blood (coagulation time 6 minutes) the mixture coagulated in 30 minutes and only a trace of prothrombin was activated. These findings suggest that the patient's blood contained an inhibitor of the platelet factor and that therefore the conversion of thromboplastinogen to active thromboplastin was prevented. This type of anticoagulant is particularly significant clinically because it cannot be counteracted by transfusion with normal blood.

Enzymatic formation and breakdown of pentose phosphate. E. RACKER (introduced by S. OCHOA). *Department of Bacteriology, New York University College of Medicine.* The oxidative decarboxylation of phosphogluconic acid to a pentose phosphate and the fermentation of the latter to CO_2 and water was previously shown to occur in yeast extracts by Warburg, Christian and Griese (Biochem. Z. **282:** 157, 1935) and by Dickens (Biochem. J. **32:** 1626, 1938). A triphosphopyridine nucleotide enzyme, oxidizing phosphogluconic acid, has now been found in bacterial extracts and has been separated by ammonium sulfate fractionation from the enzyme which oxidizes glucose-6phosphate to phosphogluconic acid (Zwischenferment). In bacterial extracts an enzyme has been found which produces triosephosphate from ribose-5phosphate. The activity of this enzyme has been measured spectrophotometrically in the presence of an excess of triosephosphate isomerase and α glycerophosphate dehydrogenase. The latter enzyme converts the triosephosphate which is formed from ribose-5phosphate, into α glycerophosphate in presence of reduced diphosphopyridine nucleotide (DPN). The oxidation of reduced DPN is followed by the disappearance of absorption at λ 340 mμ. The ribose-5phosphate splitting enzyme from bacterial extracts has been partially purified. In the same extracts an enzyme is present which oxidizes reduced DPN in the presence of glycolaldehyde. Glycolaldehyde and triosephosphate can condense to give a pentose phosphate. This reaction is catalyzed by crystalline aldolase prepared from rabbit muscle. The barium salt of the pentose phosphate has been isolated. It is not ribose-5phosphate and no evidence is available that it can be converted into ribose-5phosphate.

Enzymatic inactivation of serum vasoconstrictor. MAURICE M. RAPPORT (by invitation) ARDA ALDEN GREEN and IRVINE H. PAGE. *Research Division of the Cleveland Clinic Foundation, Cleveland, Ohio.* It is well established that perfusion of lung with serum or defibrinated blood destroys their vasoconstrictor activity. The mechanism of this destruction is probably enzymatic since a protein fraction from lung extracts inactivates highly purified preparations of serum vasoconstrictor.

Rate of protein formation in the livers of partially hepatectomized rats. D. RITTENBERG, EDITH E. SPROUL (by invitation) and DAVID SHEMIN. *Department of Biochemistry and Pathology, College of Physicians and Surgeons, Columbia University, New York.* While in a non-growing organism the rate of the synthetic reactions is identical with the rate of the degradative reactions, this cannot be the case in a growing organism. Growth results because the rate of synthesis exceeds that of degradation. Previous work from this laboratory has given values for the rate of formation of the proteins of the adult rat liver. We have now investigated the rate of protein formation in the livers of partially hepatectomized rats. Two days, after about 50% of the liver had been removed, the rats were given a standardized amount of glycine labeled with N^{15}. The rate of protein formation was measured by determining the rate of incorporation of N^{15} into the liver proteins. The data indicate that the rate of protein formation in the livers of these rats is not appreciably faster than in the livers of the normal animal. Growth in this instance is not the result of an acceleration of the synthetic mechanisms but rather appears to result from the inhibition of the degradative reactions. The implications of this concept of growth will be briefly discussed.

Determination of arginase activity in tissue

homogenates: application to epidermal carcinogenesis in mice. EUGENE ROBERTS (introduced by MICHAEL SOMOGYI). *Barnard Free Skin and Cancer Hospital, St. Louis, Mo.* Previously described methods for the determination of arginase activity have been modified to give satisfactory results with distilled water homogenates of mouse liver, kidney, epidermis, and squamous cell carcinoma. An aliquot of a 2% homogenate of the tissue was incubated with an equal volume of 2% $MnCl_2$ for 5 to 6 hours with frequent shaking. Maximal activation was achieved within 4 hours with liver, epidermis, and tumor, and was maintained through at least 6 hours of incubation. No activation of kidney homogenate was obtained with this procedure, and almost complete inactivation of the enzyme in this tissue occurred within 6 hours. The activities of suitable quantities of the homogenates before and after activation were measured in the presence of excess $MnCl_2$ by estimation of the urea formed during incubation for 10 minutes at 38°C with a concentrated arginine solution. The reaction was of zero order and the activity for each tissue was proportional to the quantity of tissue employed. Arginase activity values expressed in micrograms of urea liberated per mg of tissue nitrogen (a) before activation and (b) after activation are given below for 3 sets of tissues consisting of 10 samples each studied in epidermal carcinogenesis induced in mice by methylcholanthrene: normal epidermis, (a) 146 (61–220) and (b) 1171 (842–1659); hyperplastic epidermis, (a) 1221 (897–1682) and (b) 3074 (2,240–4380); squamous cell carcinoma, (a) 1992 (861–2390) and (b) 13,378 (5,950–18,700). These results show that a highly significant increase in arginase activity occurs in epidermal carcinogenesis.

Biochemical characterization of lymphoid tissue proteins. SIDNEY ROBERTS (by invitation) and ABRAHAM WHITE. *Department of Physiological Chemistry, Yale University, New Haven.* Mildly alkaline (pH 7.6) extracts of lymphoid tissue obtained from rabbit, mouse lymphosarcoma transplants, and calf thymus have been found to be qualitatively similar in the Tiselius apparatus. Six to eight components are evident, the quantity of each varying with species and with efficiency of extraction. Approximately 90% of the total protein present is distributed among four fractions, one of which represents more than half of the total protein in the extracts. Fractionation studies have been conducted with alkaline extracts of calf thymus. Treatment with cold ethanol yields fractions insoluble at 10%, 20%, 30%, and 40% ethanol concentrations respectively. Electrophoretic examination of these fractions suggests that a partial separation of thymus proteins has been achieved. Preliminary observations have been made on some of the chemical and physiological properties of these thymus proteins. Daily injection of the initial extract of calf thymus into the rat resulted in hypertrophy of the spleen and thymus in one to two weeks. An apparent inhibition of the normal lymphopenic effect of foreign protein injection in the rat has also been observed; this activity appeared to be characteristic chiefly of the precipitate obtained with 40% ethanol.

A fluorimetric assay for N^1-methyl-6-pyridone-3-carboxylamide. FRED ROSEN, (by invitation) W. A. PERLZWEIG, and PHILIP HANDLER. *Department of Biochemistry, Duke University School of Medicine, Durham, N. C.* Incubation of N^1-methyl-6-pyridone-3-carboxylamide (I) with acetone and KOH yields a fluorescent derivative. Traces of water inhibit the reaction. Fluorescence disappears if the final solution is acidified and may be restored by adding alkali. N^1-methylnicotinamide does not yield a fluorescent compound under these circumstances. The following procedure, while not entirely satisfactory, has been employed for urine analysis. To 10 ml. urine, adjusted to pH 9.6, 3.0 ml. saturated lead subacetate are added. After centrifugation the supernate is brought to pH 6 and evaporated in a porcelain dish. The residue is transferred to a test tube with 0.5 ml. water and washed in with about 9.5 ml. acetone. After adding 2 gm. anhydrous Na_2SO_4 and centrifuging, the supernate is evaporated and the residue extracted with a total of 8 ml. acetone. A suitable aliquot (0.2–1.0 ml.) is diluted to 1.5 ml. with acetone and 0.06 ml. 12 N KOH added. The tube is covered with parafilm and shaken for 3 hours at 20°. The solution is diluted to 10 ml. with water and read in a suitable, sensitive fluorometer using thiamine assay filters. A blank is prepared by adding 0.1 ml. water to a similar tube before the incubation period. The average 24 hour excretion of I by 6 normal adults was 8.4 mg. (5.0–13.2). After oral administration of 200 mg. nicotinamide, 150 mg. I was excreted within 24 hours and proportionally more after larger doses. Somewhat less appeared after corresponding doses of niacin. The errors of the urine extraction procedure are such that these values are probably somewhat low. Nevertheless, they establish I as the major end product of nicotinamide metabolism.

The distribution of rhodanese. OTTO ROSENTHAL. *Harrison Department of Surgical Research, Schools of Medicine, University of Pennsylvania, Philadelphia.* Rhodanese catalyzes the reaction: $HCN + Na_2S_2O_3 \rightarrow NaSCN + NaHSO_3$. Mendel, Rudney and Browman reported that the enzyme was absent in transplanted murine sarcomas and normal murine jejunal mucosa, tissues displaying high aerobic glycolysis. These authors suggested that the aerobic glycolysis was due to an inhibition of the Pasteur reaction by small amounts of HCN formed in the tissue metabolism and accumulating in the absence of rhodanese. We have determined

the rhodanese activity of H_2O homogenates of diverse normal and neoplastic tissues from different animal species. The activity was expressed in terms of μM thiocyanate/min./gm. dry weight of tissue measured at 20°C in phosphate buffer of pH 7.3, the substrate concentration being 0.05 M. The values obtained ranged from 2 to 5 in murine sarcomas and jejunal mucosa, rabbit spleen and pale skeletal muscle up to 600 in rat liver. There were pronounced species differences in the activity of homologous tissues. While our data confirm the experimental findings of the previous workers they also demonstrate that rhodanese deficiency occurs in tissues with a normal aerobic metabolism. The biological significance of rhodanese remains to be ascertained. Studies on the kinetics of the rhodanese reaction revealed apparent dissociation constants of the enzyme substrate complex of 5.5×10^{-3} and 3.8×10^{-3} for S_2O_3 and HCN respectively. The affinity of HCN to rhodanese is thus smaller than to the respiration enzyme. Under our conditions of assay 0.05 M sulfite produced a 50% reduction of the rate indicating reversibility of the reaction.

Early changes in the seminal vesicles of the castrate rat following administration of testosterone. G. G. RUDOLPH (by invitation) and L. T. SAMUELS. *Department of Biochemistry, University of Utah School of Medicine, Salt Lake City, Utah.* Fifteen and twenty hours after the subcutaneous injection of one mg. of testosterone propionate in the castrate rat there is a significant increase in the weight of the seminal vesicles. The glands have an increased water content twenty hours after injection and at this time there is an increase in the intracellular water as measured by chloride space. The oxygen uptake of the seminal vesicles increases as early as 10 hours after the injection of testosterone propionate, and this level is the same as the oxygen consumption of the glands fifteen hours and twenty hours after injection. The oxygen uptake at these times is the same as that of the seminal vesicle from a normal mature rat. When sodium succinate is used as substrate, the oxygen uptake of the seminal vesicles from castrate animals that had been injected for ten hours was increased over the oxygen uptake of the glands with glucose as substrate, while the oxygen uptake of the glands from uninjected castrate animals was only slightly increased. The changes in the glands which bring about this increased oxygen uptake will be discussed.

Oxidation of members of the Krebs cycle by liver and kidney. Inhibition by trans-aconitate. MURRAY SAFFRAN and J. L. PRADO (both introduced by K. A. C. ELLIOTT). *Department of Biochemistry, McGill University.* The addition of members of the tricarboxylic acid cycle to kidney cortex slices results in considerable acid disappearance, increased oxygen uptake, and R. Q. changes, indicating complete oxidation of these substances. This is not the case with slices of other tissues (Elliott, K. A. C., Physiol. Rev., **21**: 267 (1941)). Similar behaviour has been found with isotonic suspensions of liver and kidney cortex. The following observations were made during the course of an investigation of this difference. Low concentrations of *trans*-aconitate, an inhibitor of aconitase, depress the respiration of kidney slices in complete Ringer-bicarbonate or phosphate, but scarcely affect liver slices. In calcium-free medium, however, both are affected. The inhibitory effect is approximately proportional to the logarithm of the *trans*-aconitate concentration. Decreased respiration is found when no substrate is added, or when an excess of fumarate or malate is present, but the inhibition is overcome by the addition of succinate, citrate or *cis*-aconitate. Presumably the intense activity of the succinate-oxidizing system is not affected, and in increased concentrations, *cis*-aconitate and citrate overcome the competitive inhibition by *trans*-aconitate. In the absence of calcium, high concentrations of malate cause appreciable acid disappearance with liver slices, though the rate of oxygen uptake is actually depressed. It is tentatively concluded that both liver and kidney possess the enzymes necessary for the complete utilization of Krebs cycle acids, but that permeability factors prevent the complete oxidation of these substances by slices of tissues other than kidney.

The effects of thiamine deficiency in rats force-fed to maintain caloric balance. LEO T. SAMUELS, A. A. ANDERSON (by invitation) E. CHAPMAN (by invitation) and D. MACKAY (by invitation). *Department of Biochemistry, University of Utah School of Medicine, Salt Lake City, Utah.* To prevent the development of multiple deficiencies on thiamine-deficient diets due to the loss of appetite, rats were fed diets of constant composition by stomach tube. Both controls and deficient groups received sufficient calories to cause an increase in total weight. After 17–20 days gastric atony developed in the deficient rats. Within three days after its appearance, all these rats would be dead from gastric distention unless feeding was stopped. Consequently the rats were killed at this time and their tissues analyzed. There was no evidence of inability to form fat, both the deficient and control rats having shown a slight gain in liver fat and a marked increase in carcass fat. The nitrogen content of the deficient rats was higher and the glycogen lower than their respective controls. The kidneys of the deficient rats were 30% larger than those of the controls. Pyruvate levels in deficient rats and pyruvate oxidation were also studied.

Blood and "Tissue" protein changes in dogs on protein-deficient diets with and without sup-

plementation. GEORGE S. SAMUELSEN (by invitation), GRACE E. GRIFFIN (by invitation, LOIS E. GRIFFITH (by invitation), SAM SEIFTER and EDWARD MUNTWYLER. *Department of Biochemistry, Long Island College of Medicine, Brooklyn.* Determinations were made of the plasma volume, hematocrit, hemoglobin, total blood and plasma proteins, plasma albumin, and nitrogen balance of groups of dogs maintained respectively for three weeks on the following diets: (1) control synthetic diet containing 20% casein, (2) protein-free, (3) protein-free, choline-free, (4) protein-free plus methionine, (5) protein-free, potassium-free, and (6) a gelatin diet. Chemical studies were also made during two subsequent periods of supplementation with commercial hydrolysates of casein and lactalbumin. During depletion, the urinary nitrogen was derived mainly from sources outside of the circulation except in the case of the gelatin diet. Whereas the relative amounts of blood nitrogen and "tissue" nitrogen (nitrogen balance minus blood nitrogen) lost were reasonably constant on a given diet, their proportion varied markedly with the type of diet. Elamn's ratio of 1:30 for plasma albumin loss to total body protein loss did not hold for all diets; the protein-free plus methionine diet resulted in a ratio of 1:50, the gelatin 1:13. With nitrogen supplementation following depletion, the tissues claimed a priority for the retained nitrogen. The return to normal of circulating plasma protein did not necessarily indicate that "tissue" proteins were restored or *vice versa*. Methionine, and choline to a lesser extent, had a nitrogen sparing effect, the methionine sparing blood protein especially. Omission of potassium from the protein-free diet did not further alter nitrogen balance. Poor regeneration of blood proteins was observed after supplementation of both the potassium-free and gelatin diets.

Amino acid excretion as influenced by dietary proteins of different biological value. H. E. SAUBERLICH (by invitation) and C. A. BAUMANN. *Department of Biochemistry, College of Agriculture, University of Wisconsin, Madison, Wis.* Mice fed diets devoid of an essential amino acid are known to excrete very high percentages of all the ingested amino acids into the urine. This observation has been extended to another species, the rat, and studies were also made with mice to determine whether proteins of greater biological value than casein might not depress amino acid excretion. Rats fed diets deficient in the essential amino acids, methionine or tryptophane, excreted approximately twice the amounts of amino acids in the urine as those excreted by rats fed an adequate protein. On diets devoid of methionine or tryptophane, rats excreted 2.3% and 3.9% of the ingested tyrosine in the urine, while rats fed casein excreted only 0.7% of the ingested tyrosine in the urine. Restriction in food intake further reduced the excretion of all amino acids. Mice fed diets of a high biological value excreted less amino acids in the urine than mice fed proteins of ordinary or poor biological value, e.g. mice fed diets containing 8% of arachin, casein, fibrin, egg albumin, or lactalbumin excreted 5.0%, 4.4%, 3.3%, 1.0%, and 0.9% of the ingested histidine in the urine, respectively. The mean excretion for the seventeen amino acids determined by microbiological procedures was 4.7% for arachin, 3.4% for casein, 2.7% for fibrin, 1.5% for egg albumin, and 1.0% for lactalbumin.

Some properties of a transsulfurase responsible for conversion of cyanide to thiocyanate. J. P. SAUNDERS and W. A. HIMWICH (introduced by BERNARD J. JANDORF). *Toxicology Section, Medical Division, Army Chemical Center, Maryland.* The effect of separately varying time, temperature, pH and substrate (cyanide and thiosulfate) concentration was studied on the enzyme catalyzing the conversion of -CN to -CNS. Enzyme activity increased sharply with time of incubation until 40 minutes, from which point there was no appreciable increase with time. A sharp activity optimum was observed at 37°C. Different pH optima were shown depending upon the species from which tissue homogenates were prepared. Brain and liver tissues from dog, rat and rhesus monkey showed an optimum at pH 7.3–7.5, while tissues from rabbit gave a slightly higher optimum at pH 7.7–7.9. The pH-activity curve for rabbit tissues showed a much broader peak near the optimum than tissues from the other species examined. Maximum activity was observed when the substrate concentration was such that molar thiosulfate concentration was three times that of the cyanide present. No appreciable loss of activity was observed in homogenates stored for as long as 96 hours at 5°C, or in tissues kept for four hours at room temperature. The enzyme is inhibited by a number of compounds including S-containing compounds and -CN. If -CN is added to the tissue homogenate before addition of thiosulfate, almost complete inhibition is effected, whereas if the same amount of -CN is added after the addition of thiosulfate, normal activity is obtained. This suggests the possibility that a loose thiosulfate-enzyme combination is formed, which gives up sulfur in the presence of cyanide to form thiocyanate.

Inhibition of peptic activity by hydrazine. OTTO SCHALES, REGINA M. ROUX (by invitation), and ANNE M. SUTHON (by invitation). *Chemical Research Laboratory of the Alton Ochsner Medical Foundation and Department of Biochemistry, Tulane University, New Orleans, La.* Josephson and Euler (Ztschr. physiol. Chem. **162**: 85; 1927) observed inhibition of erepsin by phenylhydrazine, cyanide and bisulfite and concluded that this enzyme contains a group of aldehyde character.

Rona (Biochem. Ztschr. **109**: 279; 1920) found no inhibition of pepsin and trypsin by aldehyde reagents. Experiments were carried out in this laboratory in which the disappearance of the turbidity of an egg-white suspension in presence of pepsin, trypsin and papain was followed photoelectrically, using a modification of the method of Riggs and Stadie (J. biol. Chem. **150**: 463; 1943). A variety of aldehyde reagents decreased the rate of enzymatic hydrolysis of this substrate. Hydrazine was the most effective inhibitor for pepsin. Peptic hydrolysis followed a monomolecular course during the interval from 5–10 min. and the reaction constant k was a measure of pepsin activity. It was found that: (1) Two millimol hydrazine per liter decreased k to one-half its original value. (2) There was a linear relationship between k and the logarithm of the hydrazine concentration. (3) Both in the presence and absence of hydrazine there was a linear relationship between k and the reciprocal of the initial substrate concentration. (4) Graphical interpretation of experimental data indicated that the inhibition was of the non-competitive type. It is concluded that the pepsin molecule contains one or more groups which react with hydrazine and which are probably of aldehydic or closely rellated nature. The non-competitive type of inhibition suggests that these "activating" groups do not combine with the substrate during the proteolytic reaction.

Purification of phosphoglucomutase with "carbitol" acetate. Max Schlamowit (by invitation) and David M. Greenberg. *Division of Biochemistry, University of California Medical School, Berkeley.* The preparation of phosphoglucomutase from rabbit muscle with greater activity than could be gotten by previously described methods has been achieved by fractionation with "carbitol" acetate (diethylene glycol monoethylether acetate) used in conjunction with selective heat denaturation and salting-out techniques. Phosphoglucomutase isolated by the method of Colowick and Sutherland (J. Biol. Chem., **144**: 423 (1942)), which involves precipitation of the enzyme from an aqueous extract of muscle with ammonium sulfate (0.4–0.6 sat.) followed by selective heat denaturation (30 min. at 50–52° and pH 5.15), has yielded preparations of phosphoglucomutase with activities ranging up to 3,300 units per mg. protein. Phosphoglucomutase preparations with activities of 6,000 units per mg. protein could readily be obtained from aqueous muscle extract by fractionating with "carbitol" acetate (40–60% by volume). Selective heat denaturation raised the activities to around 10,000 units per mg. protein, and salting out with ammonium sulfate (0.5–0.7 sat.) further increased the activity values to 13,500–17,200 units per mg. protein. The phosphoglucomutase prepared in this manner was free from phosphorylase and hexoseisomerase activity. The enzyme unit mentioned above is defined as the γ of phosphorus of glucose-1-phosphate converted to glucose-6-phosphate phosphorus by 1 mg. of enzyme protein in 30 min. in a medium containing glucose-1-phosphate (0.162 M), Mn^{++} (1.25 mM) and CN^- (3mM) at pH 7.4.

Ribopolynucleotide fractions formed during the enzymatic hydrolysis of yeast ribonucleic acid. Gerhard Schmidt, Ricardo Cubiles (by invitation) and S. J. Thannhauser. *Boston Dispensary, Tufts College Medical School, Boston.* It was demonstrated (Cold Spring Harbor Symp., vol. 12, 1947; J. Biol. Chem., **170**: 761, 1947) that the inorganic P released by prostate phosphatase during a relatively short incubation period from ribonueleinase-treated nucleic acid originates exclusively from pyrimidine nucleotide groups. It was concluded that the action of ribonucleinase involves specifically, or at least preferentially, the pyrimidine nucleotide groups of the RNA molecule and that the enzymatic digest contains polynucleotides in which the proportion of purine to pyrimidine nucleotide groups is higher than 1:1.

The isolation of two of these polynucleotide fractions in the form of water-insoluble Ba salts has now been carried out. The action of ribonucleinase alone leads to the formation of a polynucleotide fraction in which the ratio purine nucleotide: pyrimidine nucleotide is approximately 2:1. This fraction seems to correspond to the split product described by Loring et al (J. Biol. Chem., **169**: 601, 1947) who also found that it had a high purine: pyrimidine ratio. A second polynucleotide fraction was obtained after digesting the first fraction with large amounts of prostate phosphatase. This fraction contained exclusively purine nucleotides. The polynucleotide nature of the free acids was established 1) by their low solubility in acids, 2) by their relatively very slow hydrolysis by acid phosphatase, 3) by the results of titration. These properties distinguish the polynucleotide fractions from guanylic acid which likewise forms a water-insoluble Ba salt or from mixtures of mononucleotides.

Substances isolated following the incubation of dehydroisoandrosterone, androsterone and etiocholanolone with liver tissue. John J. Schneider (by invitation) and Harold L. Mason. *Division of Biochemistry, Mayo Clinic, Rochester, Minnesota.* Dehydroisoandrosterone, androsterone and etiocholanolone were incubated as the sodium salts of their hemisuccinates with surviving rabbit liver slices for periods of three and six hours under aerobic conditions. The proteins then were precipitated with acid acetone and thoroughly washed with fresh acetone. After removal of the acetone in a vacuum the aqueous residue was made alkaline and extracted with ethylene dichloride in order to

obtain the free steroids. Further extraction after acidification of the aqueous phase gave a conjugated steroid fraction. Separation into ketonic and nonketonic fractions was accomplished with Girard's reagent T and isolation of compounds by use of chromatographic columns. Dehydroisoandrosterone was converted largely to Δ^5-androstene-3(β),17(α)-diol and in small yield to Δ^5-androstene-3(β)/16(β)/17(α)-triol. In order to study the role of the diol as a possible intermediate in the conversion of dehydroisoandrosterone to the triol, Δ^5-androstene-3(β)/17(α)-diol dihemisuccinate was incubated under the conditions indicated. Only the diol and a small amount of dehydroisoandrosterone were recovered. Compounds formed during the incubation of androsterone included androstane-3,17-dione, androstane-3(α),17(α)-diol and several as yet unidentified compounds, two of which may be isoandrosterone and androstane-3(α),17(β)-diol Etiocholanolone was converted to etiocholane-3/17-dione, etiocholane-3(α),17(α)-diol and etiocholane-3(α),17(β)-diol The significance of these results in relation to ketosteroid metabolism in general will be discussed.

Absence of a sparing action of tryptophane on nicotinamide requirements of the fly, Drosophila melanogaster. JACK SCHULTZ and GEORGE T. RUDKIN (introduced by GERRIT TOENNIES). *Lankenau Hospital Research Inst., and Inst. for Cancer Research, Philadelphia 30, Pa.* A chemically defined medium for the growth of the fly *Drosophila melanogaster*, under bacteriologically sterile conditions, has been developed (Schultz, St. Lawrence and Newmeyer, *Anat. Rec.* **96**:4/44 (1947)) This furnishes a convenient system in which effects of components of the medium on growth can be distinguished from possible effects of symbiotic microorganisms. The amino acids essential for the rat are also essential for Drosophila, as are the vitamins of the B-complex. In the work to be reported, the growth on concentrations of tryptophane from 0.06 to 0.23 mg. per ml., at levels of nicotinamide varying from 2.5 to 40 gamma per ml. medium, has been studied. Growth was measured in terms of larval length, pupation time, and weight of the adult fly. The wild type Drosophila and a vermillion-eyed mutant, which lacks the ommatin pigment of which tryptophane is a precursor, were used. In both types the nicotinamide requirements for optimum growth were found in general to increase with the tryptophane supplied. This is in apparent contradiction to the well known findings in mammals concerning the sparing action of tryptophane on nicotinamide. The contradiction is resolved by the recent finding of Ellinger and Abdel Kader (*Nature*, **160**: 675 (1947)) that the major responsibility for the nicotinamide- tryptophane interrelation rests upon the microorganisms of the gut. In the vermilion mutant, the genetic disability to use tryptophane for pigment formation lowered the tryptophane requirement for optimum growth.

Influence of the presence of a sterile abscess on the detoxication of brombenzene as mercapturic acid. JULIUS SCHULTZ (introduced by HARRY M. VARS). *Harrison Dept. of Surgical Research, Schools of Medicine, Univ. of Pennsylvania, Philadelphia.* The amount of mercapturic acid-S excreted by the rabbit following the oral administration of monobrombenzene was found to be increased in the presence of a sterile abscess. These increments were observed in both fed and fasted rabbits. In the fed rabbit receiving a single 1.5 gm. dose of brombenzene, the mercapturic acid-S was 43 and 51 mg. without a sterile abscess and 61 mg. with. In another rabbit 1.5 gm. of brombenzene on 2 successive days yielded 86 mg. of mercapturic acid-S. In the presence of a sterile abscess the excretion was 105 mg. mercapturic acid-S. During a 2 day fast the rabbits receiving 2 successive doses of brombenzene excreted 36 and 34 mg. of mercapturic acid-S. These same animals in the presence of a sterile abscess produced 42 and 54 mg. respectively. On the basis of sulfur partitions observed with these data, it can be shown that following a sterile abscess there was an increase in neutral sulfur. It appears that this neutral sulfur may be utilized by the animal in forming mercapturic acid. These data indicate that a specific metabolic reaction can utilize the tissue breakdown products resulting from the formation of a sterile abscess. The significance of these findings in relation to the protective action of a sterile abscess against chloroform poisoning will be discussed.

Studies on folic acid conjugase in blood. B. S. SCHWEIGERT (introduced by P. B. PEARSON.) *Dept. of Biochemistry and Nutrition, A & M College of Texas, College Station.* In the course of investigations on the influence of the dietary intake pteroylglutamic acid (folic acid) on the amount present in blood before and after enzymatic treatment, the occurrence of folic acid conjugase in blood has been demonstrated. Microbiological analyses on samples of whole blood or plasma (turkey) incubated at pH 7.0 for 16–18 hours were 5–20 times higher than analyses made on similar samples prior to incubation. Separate studies showed that greater liberation of the vitamin occurred when the blood was incubated at pH 7.0 or 8.0 than at pH 4.5 or 6.0. A further increase in the amount liberated was obtained when the samples were incubated with a chick pancreas enzyme preparation (pH 7.0). Preliminary evidence indicates that the concentration of folic acid in blood determined either before or after enzymatic treatment (chick pancreas enzyme) is influenced by the amount of the vitamin ingested by the turkey.

Turkeys maintained on a purified ration deficient in pteroylglutamic acid showed poor feathering, a slow rate of growth and a low hematocrit as well as a low concentration of pteroylglutamic acid in the blood.

Purification of alpha-amylase from barley malt extracts. SIGMUND SCHWIMMER. *Enzyme Research Laboratory, Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, USDA, Albany, California.* The Maltose Equivalent of purified enzyme preparations has been raised from 350,000 as previously reported (Cereal Chem., **24,** 315 (1947)), to 430,000 units of alpha-amylase per mg. of protein. The new preparations were made from a commercially obtained concentrated barley malt extract instead of whole malt. This extract contains smaller amounts of the less soluble protein impurities which are present in usual grain extracts. Adsorption of the enzyme on bentonite was no longer required and purification was achieved merely by a series of ammonium sulfate fractionations of the heated extract. The less soluble protein impurities present at later stages of purification include an iron-rich globulin which apparently forms a solid solution with the enzyme protein on precipitation. After each fractionation step, some of the less soluble impurity can be removed by dialysis against saturated calcium sulphate solution. The more soluble protein impurities remain in the supernatant of the precipitated enzyme solution. Preliminary ultracentrifuge measurements on a preparation so pruified indicate that the bulk of the protein present sediments as one component.

Bacteriological characteristics of mandelamine. JOHN V. SCUDI and CHARLES J. DUCA (by invitation). *Nepera Chemical Co. Inc., Yonkers, N. Y.* Mandelamine (methenamine mandelate) was compared with Streptomycin and with Sulfathiazole. Each drug was dissolved in sterile, buffered urine using a serial two-fold dilution technique, and 17 strains of bacteria commonly associated with urinary tract infections were then challenged with each of the three drugs. Bacteriostatic and bactericidal studies showed that Mandelamine compares very favorably with Streptomycin and with Sulfathiazole in activity. Minimal bacteriostatic concentrations of Mandelamine were generally the same as its bactericidal concentrations. Lowering the urinary pH from 6.5 to 5.5 increased the activity of Mandelamine. Resistance to Streptomycin and Sulfathizaole appeared rapidly and to a marked degree. Organisms rendered resistant to these drugs at one pH remained resistant at higher pH levels. Further, organisms rendered resistant to these drugs remained fully susceptible to Mandelamine. In sharp contrast with the results obtained with Streptomycin and Sulfathiazole, it was impossible to elicit any direct evidence of resistance to Mandelamine. The clinical implications of these findings are discussed.

Pharmacological characteristics of neohetramine, a new antihistaminic drug. JOHN V. SCUDI, JOHN F. REINHARD (by invitation) and N. B. DREYER (by invitation). *Nepera Chemical Co., Inc., Yonkers, N. Y. and the Department of Pharmacology, University of Vermont.* Neohetramine, 2 - (N - dimethylaminoethyl - N - p - methoxybenzyl) aminopyrimidine monohydrochloride, is relatively non-toxic and is efficient both in counteracting the effects of histamine and in preventing anaphylactic shock. The acute toxicity of Neohetramine in mice is about one-half that of other antihistaminics. Weanling rats receiving as much as 200 mg/kg. of Neohetramine daily for 3 months grew at a normal rate, presented no abnormalities in blood morphology and developed no organ pathology. Neohetramine showed marked activity against the bronchiolar, capillary, local vasodilator, smooth muscle and vasodepressor actions of histamine, but the number of molecules of histamine antagonized by 1 molecule of Neohetramine varied from 0.001 to 9 depending on the species of animal and test procedure. Neohetramine protected against anaphylactic shock in actively and passively sensitized guinea pigs but the effective dosage was 5 to 20 times that required for comparable protection against histamine shock.

The general pharmacologic actions of Neohetramine are as follows: on smooth muscle, low concentrations are depressant, large concentrations induce contraction. In the eye, it produces a transient irritation accompanied by local anesthesia comparable to that obtained with procaine. It does not alter sympathetic responses, nor does it potentiate the action of epinephrine. Neohetramine enhances the intestinal response to vagal stimulation but it is also weakly parasympatholytic, causing slight depression of salivary secretion induced by stimulation of the chorda tympani nerve.

Purification of prothrombin and thrombin. WALTER H. SEEGERS and ARNOLD G. WARE (by invitation). *Department of Physiology, Wayne University, College of Medicine.* The purification procedures described for obtaining highly active prothrombin (Arch. Biochem. **6,** 85, 1945) have been subjected to extensive study. The former work has been repeated approximately 200 times with various modifications and it is believed that approximately the methods previously described give the highest purification thus far achieved. Thrombin of purity equivalent to that described by Seegers and McGinty (J. Biol. Chem. **146,** 511, 1942) has also been obtained. Products of this kind were examined for us in the ultracentrifuge by Gerson Kegeles and J. W. Williams of the University of Wisconsin. Both proteins were found to be polydisperse. The prothrombin molecule (ap-

proximately 140,000) is approximately twice the size of the thrombin molecule (approximately 77,000). In recent work purified prothrombin has been obtained in stable form. This is accomplished by the simple expedient of storing the $Mg(OH)_2$ eluate, described on page 88, Arch. Biochem. **6**, 1945, at 5°C. overnight instead of storing it at room temperature. This maneuver precludes the appearance of small amounts of thrombin in the final product. Small amounts of thrombin destroy prothrombin. It is estimated that the prothrombin product contains less than 1% Ac-globulin as impurity. This Ac-globulin activity can be destroyed by heating the prothrombin at 53°C. for 2 hours.

A quantitative method for a serum polysaccharide present in cases of tuberculosis and carcinoma. F. B. SEIBERT, M. L. PFAFF (by invitation), and M. V. SEIBERT (by invitation). *Henry Phipps Institute, University of Pennsylvania, Philadelphia, Pa.* A simple method useful for routine clinical analyses, involving the reaction between a carbohydrate complex in the serum and tryptophane in the presence of perchloric acid, has been utilized to detect the presence of a substance which appears in the sera of such diseases as tuberculosis and carcinoma of several types. The nature of this substance has been considered at length and it was finally suggested, largely on spectrophotometric data, that it might consist of a fructose compound or the nucleic acids and their hydrolysis products or derivatives. Very little, if any, dialyzes away from normal serum whereas a considerable proportion diffuses from the pathological sera, and the presence of nucleic acid could not be established in these diffusates. Further work is needed to establish its identity.

A statistically significant increase over the normal value was found even in cases of minimal active tuberculosis and as the disease progressed to moderately and far advanced stages, this substance progressively increased, in some cases to more than 200 per cent. The mean value for minimal cases which have become of questionable clinical significance is the same as for normals but that for the moderately advanced cases in this category is increased.

Concentration changes of certain vitamins and enzymes in livers of rats on protein-free diet SAM SEIFTER, DAVID M. HARKNESS (by invitation), B. FELDMAN (by invitation), LEONARD RUBIN (by invitation) and EDWARD MUNTWYLER. *Department of Biochemistry, Long Island College of Medicine, Brooklyn.* The present report initiates an investigation into the possible relationships existing among enzyme activities, coenzyme concentrations, and protein levels of livers of animals on restricted protein intake. The effect of protein depletion was determined on (1) an enzyme having no known vitamin coenzyme (arginase), (2) an enzyme having a riboflavin coenzyme (d-amino acid oxidase), and (3) nicotinic acid and riboflavin, which are active in numerous enzyme systems. Later studies will deal with the coenzyme-enzyme-protein relationships. Three groups of Wistar albino male rats between 16 and 25 weeks of age were placed on a protein-free diet, adequate in all other respects, for periods of one, two, and three weeks respectively. Pair-fed controls were maintained concurrently on an adequate diet containing 20% casein. At the end of the feeding periods the animals were sacrificed without anesthesia and the livers analyzed for water, fat, nitrogen, riboflavin and nicotinic acid (microbiologically), d-amino acid oxidase (by pyruvate formation from alanine), and arginase (by urea formation from arginine).

Animals on the protein-free diet showed an increase in liver fat and a decrease in liver nitrogen as compared with the pair-fed controls, and the changes were proportional to the duration of the depletion period. The livers of the depleted animals also showed a decrease in nicotinic acid, riboflavin, d-amino acid oxidase, and arginase as compared with the controls; all changes were proportional to the decrease in hepatic nitrogen.

Lipotropic activity and toxicity of methoxinine (oxy-methionine). C. BOYD SHAFFER and FRANCES H. CRITCHFIELD (introduced by LEONARD H. CRETCHER). *Chemical Hygiene Fellowship, Mellon Institute, Pittsburgh, Pennsylvania.* Four groups of 10 rats each were fed a lipogenic diet for a period of 21 days. The animals initially weighed between 130–180 gms., with equal numbers of males and females per group. The basal diet consisted of lard 40, casein 5, sucrose 46, salts 4, and cellulose 5. Purified vitamins were supplied in tablet form. In addition, the following supplements were given daily by stomach tube in water solution: Group II, 50 mg. methoxinine; Group III, 50 mg. methionine; Group IV, 50 mg. methoxinine plus 50 mg. methionine. Group I served as controls. All animals in Group II died between the 12th and 20th day, and are therefore excluded from further comparison. Those in the other groups lost weight rapidly, but all survived the experimental period. Liver and kidneys were weighed at sacrifice, and liver lipid (fatty acids plus cholesterol) determined by a micro-oxidative method. This and other pertinent data are tabulated below.

	Group I	Group III	Group IV
Food intake, gm./rat/day	4.16	4.11	4.24
Mean wt. loss (% of initial wt.)	16.60	13.20	34.40*
Liver wt. (% of body wt.)	6.10	6.33	5.13*
Kidney wt. (% of body wt.)	0.87	0.92	1.08*
Liver lipid (% wet organ wt.)	19.90	14.90*	7.02†

* Statistically significantly different from Group I.
† Statistically significantly different from Groups I and III.

From this it is concluded that methoxinine exerted a definite lipotropic effect, in addition to an inherent toxic action.

The in vitro synthesis of heme from glycine by the nucleated red blood cell. DAVID SHEMIN, IRVING M. LONDON (by invitation) and D. RITTENBERG. *Departments of Biochemistry and Medicine, College of Physicians and Surgeons, Columbia University, New York.* It has been demonstrated, in the intact mammal, that glycine is the nitrogenous precursor of the protoporphyrin of hemoglobin. For further investigation of the mechanism of porphyrin formation, we sought a simpler biological system. We have found that whole blood of the duck can form heme from glycine *in vitro*. On aerobic incubation of whole duck blood with glycine labeled with 32 per cent N^{15}, isotopically labeled heme was synthesized. After four, seventeen and twenty-four hours the N^{15} concentrations of the isolated hemin were 0.051, 0.108 and 0.113 atom per cent excess N^{15} respectively. Further work is now in progress to determine the optimal conditions for the *in vitro* synthesis of heme, to determine the metabolites other than glycine which are necessary for this synthesis, and to study the chemical mechanisms and the enzymatic systems involved in this synthesis. In addition the use of this *in vitro* system is being extended to the investigation of protein and nucleoprotein synthesis. We had earlier found that the synthesis of heme occurs at an appreciable rate with the blood of patients with sickle cell anemia.

Nature of the "sporogenes vitamin" and nutrition of clostridium sporogenes. G. M. SHULL (by invitation), RICHARD W. THOMA (by invitation), and W. H. PETERSON. *Department of Biochemistry, University of Wisconsin, Madison.* The so-called "sporogenes vitamin" of Fildes, Pappenheimer, et al., is a growth factor required by *Clostridium sporogenes*. Moderate growth of eight strains of this organism has been obtained on a chemically defined medium containing amino acids, glucose, biotin, PAB, nicotinic acid, salts, buffer, and sodium thioglycolate. For heavy growth an unidentified factor, probably a polypeptide, is particularly effective. However, the unknown factor can be replaced by rather large amounts (about 25 mg. per 10 ml.) of arginine in combination with either tyrosine, phenylalanine or tryptophane (e.g. 20 mg. per 10 ml.). These amino acids appear to be preferential substrates, since they are metabolized with the production of ammonia even in the presence of glucose. Biotin is the only vitamin for which an absolute requirement exists. PAB and nicotinic acid exert a mild stimulatory effect on the growth of the organism. Oxybiotin can replace biotin but is required in about twice the concentration. Oleic, linoleic, ricinoleic, and vaccenic acids are active, in the order named, in replacing biotin both in the basal medium and in an arginine-tyrosine medium. The use of emulsifying agents such as the Tweens greatly facilitates the demonstration of this phenomenon. Tween 80, an oleic acid ester, is active in high concentrations in replacing biotin.

It thus appears probable that either biotin or an unsaturated fatty acid, or both, were components of Pappenheimer's concentrates of the "sporogenes vitamin".

The urinary excretion of amino acids and peptides after intravenous infusion to dogs. ROBERT H. SILBER, E. E. HOWE, and CURT C. PORTER (introduced by EDGAR G. MILLER). *Merck Institute for Therapeutic Research and the Merck Research Laboratories, Rahway, N. J.* Four protein hydrolysates and one amino acid mixture prepared in 5 different laboratories were given to dogs intravenously as the sole source of protein nitrogen daily for a period of 4 days, at a rate of 2 mg. N per kg. per min. and a total dose of 120 mg. N per kg. Only 5.5% of the amino acids in the amino acid mixture was lost in the urine whereas 15 to 29% of the hydrolysates was excreted in the form of amino acids and peptides. The amino acids of 3 protein hydrolysates were retained more efficiently than the peptides, with only 10 to 15% of the free amino acids appearing in the urine in contrast to 29 to 45% of the peptides. These hydrolysates contained 25 to 45% of their amino acids in peptide form.

The pattern of essential amino acids excreted in the urine after infusion of a peptide-free amino acid mixture was compared with the pattern of amino acids infused (6 mg. N per kg. per min. and a total dose of 200 mg. N per kg.). The pattern of the infused mixture was then altered on the hypothesis that those amino acids which were excreted in disproportionately large amounts were present in excessive amounts in the mixture, and conversely, that those excreted in disproportionately small amounts were present in too low a concentration in the mixture. It has been possible in this way to obtain an amino acid mixture which more closely resembles the pattern of amino acids excreted in the urine. The significance of this will be discussed.

The effect of hypertrophy on the chemical composition of rat cardiac muscle. LEONARD T. SKEGGS (by invitation), JACK R. LEONARDS (by invitation), and VICTOR C. MYERS. *Department of Clinical Biochemistry, Western Reserve University, Cleveland, Ohio.* The purpose of the present study was to determine the effect of cardiac hypertrophy on those chemical constituents which are known to play a major role in muscle contraction and also to evaluate chemically the presence of anoxia of the heart muscle. Cardiac hypertrophy was produced in rats by a variety of accepted

methods including hypertension, anemia and excessive administration of desoxycorticosterone acetate. These procedures increased the heart weight to body weight ratio by 40 to 75 per cent above normal. The hearts were then analyzed for inorganic phosphorus, phosphocreatine, the easily hydrolyzable phosphorus of adenosinetriphosphate and adenosinediphosphate, total acid soluble phosphorus, glycogen, and lactic acid. The presence of cardiac hypertrophy did not significantly change the concentration of any of the above mentioned constituents with the exception of the anemic animals in which there occurred a slight increase in lactic acid. If the cardiac muscle in hypertrophy was in a state of anoxia one would expect to find a marked increase in lactic acid and a fall in phosphocreatine and adenosinetriphosphate. Such changes were actually observed in the hearts of normal animals subjected to an atmosphere of 3 per cent oxygen for 5 minutes. It therefore appears that hypertrophy can exist in rat hearts without the accompaniment of detectable chemical changes. However, it is realized that marked changes may occur in the presence of a greater degree of hypertrophy or during heart failure and this phase is now being studied.

The hydrolysis of glycylglycine and glycyl-L-leucine by peptidases. EMIL L. SMITH. *Laboratory for the Study of Hereditary and Metabolic Disorders, and the Depts. of Biochemistry and Medicine, University of Utah School of Medicine, Salt Lake City, Utah.* Although many dipeptides are hydrolyzed by animal tissues, the specificity and nature of the responsible enzymes have remained for the most part unknown. The hydrolysis of glycylglycine (GG) by extracts of rat muscle and human uterus is due to a specific dipeptidase which is strongly activated by Co^{++}, and to a lesser extent by Mn^{++}, but not by other divalent ions. This enzyme does not hydrolyze the tripeptide, glycylglycylglycine, or substituted compounds like benzoylglycylglycine, carbobenzoxyglycylglycinamide, benzoylglycinamide, glycylglycinamide, or glycylsarcosine. Sarcosylglycine is hydrolyzed although more slowly than GG. The hydrolysis of GG does not appear to parallel the hydrolysis of other dipeptides. The hydrolysis of GG follows the kinetics of a zero-order reaction. The maximal activity is at pH 7.6. The apparent dissociation constant of the cobalt-enzyme compound is 2.8×10^{-5} molar.

The glycyl-L-leucine (GL) hydrolyzing enzyme of rat muscle is activated by Mn^{++}, that of human uterus by Zn^{++}. Maximal activity in both cases is obtained with phosphate buffer apparently because of the removal of Ca^{++} which acts as an inhibitor. With activating metal and phosphate, the hydrolysis of GL is a first order reaction, and is proportional to the enzyme concentration over a wide range. The enzyme possesses the specificity of a dipeptidase since carbobenzoxyglycyl-L-leucine, carbobenzoxyglycyl-L-leucinamide, and glycyl-L-leucinamide are hydrolyzed either not at all or only very slowly by extracts rich in GL activity.

The riboflavin requirement of the normal infant. SELMA E. SNYDERMAN (by invitation) KATHERINE C. KETRON (by invitation), ANTHONY A. ALBANESE and L. EMMETT HOLT, JR. *From the Department of Pediatrics, New York University and the Children's Medical Service of Bellevue Hospital, New York City.* In determining the minimal requirement of the infant for riboflavin, it was not thought justifiable to induce symptoms of this deficiency. Instead, the attempt was made to ascertain accurately the riboflavin intake which would lead to minimal urinary excretion values. It has been clearly demonstrated in the case of thiamine, that when the intake is gradually reduced, the urinary excretion falls to a minimal level which is first reached at an intake two to three times as great as that which produces symptoms There is evidence that a similar relationship occurs in the case of riboflavin. Infants were placed on metabolism frames and given purified diets in which practically all the riboflavin was provided as a weighed supplement. By gradual adjustment of the riboflavin ingestion over a period of months, the riboflavin intake that would just remain the minimal urinary excretion without additional spilling was determined. Maintenance at this level for many weeks produced no manifestations of riboflavin deficiency. This level, 0.40 to 0.45 mg. per day would therefore seem to be an adequate one and presumably not far above the critical value. This intake was also checked by measurements of the riboflavin of the blood serum, the red blood cells, and the white blood cells. Only when the intake was dropped below the point of minimal urinary excretion were low blood values observed.

Vitamin A absorption in newborns, older children, adults, and a storage in rats. ALBERT E. SOBEL, LOTTIE BESMAN (by invitation) and BENJAMIN KRAMER. *Departments of Biochemistry and Pediatrics, The Jewish Hospital of Brooklyn,* **16,** *N. Y.* Newborn children show impaired intestinal absorption compared to children above one year of age or adults, as indicated by low vitamin absorption curves after feeding Oleum Percomorphum (Vitamin A ester) or the nonsaponifiable fraction of fish liver oil (Vitamin A alcohol) dissolved in maize oil. On feeding Vifort, which is an aqueous dispersion of Vitamin A alcohol (containing the vitamin B complex, Vitamins C and D) elevated vitamin A absorption curves were observed. Upon administering an aqueous dispersion of vitamin A similar to Vifort, containing the same amount of dispersing agent (16% sorbitan monolaurate polyoxalkalene derivative) but no other

vitamins a rise similar to that found with Vifort was obtained. *Thus it appears that in newborn children there is an impaired absorption of Vitamin A in oil,* which may be remedied when the vitamin A is given in an aqueous dispersion. Moreover, the % of dispersing agent and not the absence of vitamin supplements is the important factor in determining the magnitude of improved absorption. In older children and in adults there was also improved absorption with aqueous dispersions as measured by vitamin A tolerance curves.

Liver storage in rats was about 3 times as high with vitamin A in aqueous media (with or without supplements) than the same amount of vitamin A dissolved in oil.

Influence of feeding amino acids on the spinal fluid level of free amino acids. JAMES D. SOLOMON and STANLEY W. HIER (introduced by OLAF BERGUIN). *Department of Biological Chemistry, University of Illinois College of Medicine and the Research Laboratories, The Wilson Laboratories, Chicago, Ill.* In a preceding abstract the levels of eleven individual free amino acids in cerebrospinal fluid were reported. The results of some experimentally produced changes in the concentrations of arginine, histidine, isoleucine, leucine, lysine, phenylalanine, threonine, tyrosine, valine, methionine, and cystine in the spinal fluid of human subjects are reported. The amino acids were fed orally in aqueous suspension 4 hours before collecting the spinal fluid by lumbar puncture. All were fed in combination with dl-isoleucine and in addition some were fed singly. It was found that there was a considerable variation in the rise of single amino acid from the average normal level. It was also observed that the effect of dl-isoleucine on the rise of other amino acids when fed in combination showed maa rked difference. For example, 25 grams of l(+)arginine given with 15 grams of dl-isoleucine caused arginine to rise 80% and isoleucine to rise 311% above the average normal values. On the other hand, 25 grams of l(+)histidine given with 15 grams of dl-isoleucine caused a rise in the histidine value to 304 per cent and the isoleucine value to 89% above the average normal values. When 25 grams of dl-methionine were given alone (12 hours before collection of sample) there was a rise of 325 per cent, while the ingestion of 25 grams of dl-valine alone (4 hours before collection of sample) the rise in the valine level was 900 per cent above the average normal level.

A paradoxical effect of insulin on glucose assimilation. MICHAEL SOMOGYI. *From the Laboratory of the Jewish Hospital of St. Louis, St. Louis, Mo.* Insulin action is generally associated with an acceleration of glucose assimilation by the tissues (transfer of blood sugar into tissue cells). If a healthy person receives 50 gm. of glucose by mouth, a *simultaneously* administered small dose of insulin (2.5–5 units) increases the rate of assimilation, both in the liver and in the peripheral tissues. This is reflected by the fact that the glucose-time (glucose tolerance) curve will run on much lower levels than if glucose alone is administered (Himsworth). Our experiments are in line with this known fact. If, however, the same small dose of insulin is given during the postabsorptive state and is allowed to produce hypoglycemia, the assimilation of subsequently administered glucose is greatly inhibited instead of being accelerated. This fact was demonstrated in our experiments as follows: Healthy adult persons were given intravenously 4–5 units of crystalline insulin, and 30–60 minutes later were fed 50–100 gm. of glucose by mouth. Although at the moment of the ingestion of the glucose the blood sugar was at hypoglycemic levels, it invariably rose to much higher hyperglycemic levels than if the same amount of glucose was fed without the administration of insulin. Indeed, in some instances the hyperglycemia reached *diabetic levels*. This phenomenon is interpreted as follows: The hypoglycemia, in the present instance caused by insulin, mobilizes (activates) an insulin-antagonistic mechanism which inhibits insulin action. This mechanism (the identity of which we shall not discuss at this occasion) does not stop at simply nullifying the action of the injected insulin, but—by over-compensation—exerts a "diabetogenic" effect. Our clinical studies proved that recognition of this phenomenon has a far-reaching significance in the insulin treatment of diabetes mellitus.

An enzymatic micromethod for determination of acetic acid. MORRIS SOODAK (by invitation) and FRITZ LIPMANN. *Biochemical Research Laby., Massachusetts General Hospital, and the Departments of Biological Chemistry and Medicine, Harvard School, Boston.*—In enzymatic acetylation of sulfanilamide in pigeon liver extracts, acetate may be made the limiting factor, cf. Kaplan and Lipmann, Federation proceedings **6**, 266, 1947. This system may be adapted to quantitative determination of small amounts of acetate. For such purpose, it is necessary to use a purified enzyme. From acetone powder extract, an active preparation was obtained between 40 and 70 per cent saturation with ammonium sulfate. Care must be taken to eliminate traces of acetate usually present in ATP. Barium nitrate has to be used as precipitant for ATP-preparations used in this method. In such a system, between 3 and 12 μg of acetate per 0.3 ml., the acetylation of sulfanilamide is proportional with the acetate concentration. In over hundred-fold concentration, propionate, butyrate, and valerate interfere with acetate determination.

A very interesting interference was observed with acetoacetate. It could be shown that the activity of acetoacetate is due to its preliminary

attack by an independent enzyme. Some application of the method in the study of acetate metabolism will be reported.

The determination of choline in brain lipids. WARREN M. SPERRY and FLORENCE C. BRAND (by invitation). *Dept. of Biochemistry, New York State Psychiatric Inst., New York.* For the application of our micro-method (Fed. Proc., **4**, 104, 1945) to the determination of choline in brain lipids it was necessary to find an agent which would hydrolyze sphingomyelin and which could be removed, or treated so it would not interfere in the procedure. HI gave promise of satisfying these requirements, but the occasional low recoveries of choline, noted previously (Fed. Proc. **5**, 155, 1946), were found to represent destruction of choline by HI. Glacial acetic acid appeared to cleave choline quantitatively from brain lipids, but the reaction is slow, requiring long heating in sealed tubes. Dilute HNO_3, suggested by Ducet and Kahane (Bull. soc. chim. biol. **28**, 794, 1946), did not yield satisfactory results. With a saturated $Ba(OH)_2$ solution there were large losses which were traced mainly to hydrolysis in glass apparatus and were presumably caused by traces of silica. Hydrolysis in monel tubes greatly improved the results, but recoveries were still about 10 to 15% low. The loss did not seem to represent a destruction of choline as it was more or less independent of the time of heating. When choline was heated in a half-saturated $Ba(OH)_2$ solution, found by Dr. Jordi Folch-Pi to cleave choline from lipids (personal communication), there was still a small apparent loss (about 5%), but this can be corrected by suitable controls. Half-saturated $Ba(OH)_2$ appears to satisfy reasonably well the requirements stated above, and to make possible the determination of choline in brain lipids with our method.

Polarographic studies in cerebrospinal fluids. MONA SPIEGEL-ADOLF and ARNOLD S. J. LEE (by invitation). *Dept. of Colloid Chemistry, Temple University School of Medicine, Philadelphia, Penna.* In a first series of experiments polarography was used for the study of some organic substances in the cerebrospinal fluid (CSF). Former studies (J. Nerv. Ment. Disease **89**, 311, 1939) had ascertained quantitative and qualitative changes in pathologic CSF's escaping routine methods. A self recording polarograph (Leeds and Northrup) was available. In preliminary experiments the findings of Gillam about the polarographic behavior of ascorbic acid could be confirmed. As little as 0.3 mg./100 ml. can be detected by this procedure, both in artificial mixtures sufficiently similar to CSF and in genuine CSFs. The presence of nucleic acids and derivatives does not appreciably influence these results. For the simultaneous determinations of ascorbic acid and nucleic acid and derivatives a combination of polarographic, ultraspectrographic (D U Beckman photoelectric Quartz spectrophotometer) and chemical methods was used. It could be shown that the selective absorption at about 265 mμ is due to both nucleic and ascorbic acid as has been assumed previously by one of us (J. Phys. Chem. **50**, 447, 1946). After the oxidation of ascorbic acid a part of the selective absorption subsides. Through colorimetric studies the presence of nucleic acid derivatives in the CSF of pathologic origin could be confirmed. In a final series of studies the colloidal power of CSF in suppressing the initial peak of lead chloride has been tested. Experiments are in progress to correlate these findings to the protein content and gold sol reaction of the CSFs.

The metabolism of the α, γ and β hydrogen atoms of L-leucine. DAVID B. SPRINSON (by invitation) and D. RITTENBERG. *Department of Biochemistry, College of Physicians and Surgeons, Columbia University, New York.* The occurrence, in mammalian tissues, of an enzyme system capable of hydrolyzing dehydro peptides has been considered to support the view that dehydrogenation of proteins or peptides is a significant metabolic reaction. In order to investigate this possibility α, β, γ-deutero-L-leucine containing N^{15} in the amino group was synthesized and fed to rats. L-leucine was then isolated from the body tissues. By appropriate degradative methods the concentration of deuterium at each of the labeled positions was determined in the compound fed and the one isolated. It was then possible to calculate the changes in $D:N^{15}$ ratio for the α, β, and γ positions of the amino acid following interaction with the tissue proteins. The results indicate that the β and γ deuterium atoms are diluted about one-half as rapidly as the N^{15}. The α deuterium atom, on the other hand, was removed three times as fast as the nitrogen. It, therefore, appears that the leucine could not have undergone extensive α, β-dehydrogenation. The extraordinary lability of the α-hydrogen may be due to the influence of transaminase enzyme systems such as recently discussed by Konikova, Dobbert and Braunstein (Nature, **159**, 67 (1947)).

The concentration of the labile factor of prothrombin in the blood of various species. MARIO STEFANINI (by invitation) and ARMAND J. QUICK. *Department of Biochemistry, Marquette University School of Medicine, Milwaukee.* When oxalated human plasma is stored, prothrombin activity decreases due to the disappearance of a plasma principle, which was originally named component A, but which has been renamed the labile factor. Since this principle is not adsorbed by $Ca_3(PO_4)_2$, plasma treated with this reagent retains its labile factor, but loses its prothrombin activity. When this plasma is added to stored plasma, it reduces

the prothrombin time to normal or even shorter. On this finding a quantitative test was developed for estimating the concentration of the labile factor in plasmas of different species. Oxalated human plasma was stored until its prothrombin time exceeded 35 seconds. The degree of delay does not significantly influence the test. The plasma to be tested was treated with $Ca_3(PO_4)_2$. One volume of this plasma was added to 9 volumes of stored human plasma and the prothrombin time determined. Since human plasma was found to contain the smallest concentration of the labile factor, it was made the standard for comparison. The plasmas from other species were diluted until they approximated the concentration of the labile factor in human plasma. The degree of dilution served as a measure of the relative concentration. On this basis, rabbit plasma contains 50, dog 10, cow and guinea pig 5 times as high a concentration as human plasma. It appears that the rapid loss of prothrombin activity of human plasma is attributable to its low content of the labile factor.

Mechanism of hyperbilirubinemia due to sodium nicotinate. MARIO STEFANINI (introduced by A. J. QUICK). *Department of Internal Medicine, University of Roma, Italy and Department of Biochemistry, Marquette University School of Medicine, Milwaukee, Wisconsin.* It has been found (Am. J. Med. Sc., **213,** 150, 1947) that the transitory rise of indirect reacting bilirubin of serum following intravenous injection of 30 mg. of sodium nicotinate intravenously in man is not due to exaggerated hemocatheresis or to mobilization of tissular deposits of biliary pigments. The problem was further studied by injecting sodium nicotinate in normal subjects while collecting as much bile as possible for two hours with continuous suction through a Rehfuss tube passed into the duodenum. The injection was followed by a remarkably copious flow of bile for about one hour. The serum bilirubin increased much less than when bile was not removed; this agrees with the finding that sodium nicotinate causes a minimal rise of serum indirect reacting bilirubin in patients with complete biliary obstruction. In biliary obstruction or when bile was removed from the duodenum, the transitory increase in urobilin excretion normally occurring within two hours after injecting sodium nicotinate was not observed. These experiments suggest that: 1) sodium nicotinate injection is followed by hyperbilirubinemia only if bile is normally present in the intestine. The substance apparently stimulates the bile flow thus increasing the quantity of mesobilirubinogen available for reabsorption in the blood stream through the venous extraportal system (Blankerhorn, J. Exp. Med., **45,** 195, 1927) as indirect reacting bilirubin. 2) mesobilirubinogen, serum indirect reacting bilirubin and urobilin are strictly related. 3) increased urobilinuria may not always be due to pathological conditions but may result from changes in absorption of mesobilirubinogen.

Chromatography of amino acids. Solvent systems for the fractionation of protein hydrolysates. WILLIAM H. STEIN and STANFORD MOORE (by invitation). *Rockefeller Institute for Medical Research, New York City.* Employing potato starch columns (cf. Elsden and Synge, Biochem. J., **38,** Proc. IX, 1944; Synge, Biochem. J., **38,** 285, 1944), with the solvent system 1:1 butanol:benzyl alcohol-water, quantitative chromatographic methods have been developed for phenylalanine, leucine, isoleucine, tyrosine, and valine in protein hydrolysates (Moore and Stein, Annals N. Y. Acad. Sciences, in press). Methionine has subsequently been rendered quantitative in this solvent. By the use of other solvents the technique has been extended to include additional members of the amino acid series. With a solvent system which gives more rapid band rates, a quantitative picture can be obtained which includes all of the amino acid components of a protein hydrolysate, with a few overlaps. For example, a single chromatogram (0.9 x 30 cm.) run with 1:2:1 *n*-butanol:*n*-propanol:0.1 N HCl followed (after the emergence of aspartic acid) by 2:1 *n*-propanol:0.5 N HCl gives successive effluent peaks corresponding to the following components: leucine-isoleucine, phenylalanine, methionine-valine, tyrosine, proline, glutamic acid-alanine, threonine, aspartic acid, serine, glycine, ammonia, arginine, lysine, histidine, and cystine. This type of experiment has been used for screening work and has given quantitative values for those components which emerge as single peaks. The procedure has been simplified by the use of narrow columns about 1 cm. in diameter. One-half cc. fractions are collected directly in colorimeter tubes on an automatic fraction collector and the entire fraction submitted to colorimetric ninhydrin analysis, thereby eliminating the pipetting of aliquots. About 2.5 mg. of protein are required per chromaogram.

Synthesis of S-benzyl-thiopyruvic acid and its conversion to N-acetyl-S-benzyl-L-cysteine in the rat. JAKOB A. STEKOL. *The Lankenau Hospital Research Institute and the Institute for Cancer Research, Philadelphia, Pa.* S-Benzyl-thiopyruvic acid was synthesized from chloropyruvic acid and benzyl mercaptan, and characterized. One per cent of S-benzyl-thiopyruvic acid mixed with a complete casein diet was fed to four adult rats, and from the urine of these rats N-acetyl-S-benzyl-L-cysteine was isolated and identified by analysis and by comparison with the synthetic material and with a similar product which was excreted by the rats on administration of benzyl chloride or S-benzyl-L-cysteine. The results are proof for the conversion of S-benzyl-thiopyruvic acid to the

corresponding N-acetyl-L-amino acid derivative *in vivo*. The data presented, together with the earlier work, offer experimental evidence for the hypothesis expressed in regard to the possible formation of S-benzyl-thiopyruvic acid as the intermediate in the inversion of S-benzyl-D-cysteine to N-acetyl-S-benzyl-L-cysteine *in vivo* (J. Biol. Chem., **124,** 129 (1938); **128,** 199 (1939)).

Influence of the thyroid upon incorporation of deuterium into tissue constituents of the rat. DEWITT STETTEN, JR., and ADELE KARP (by invitation). *Department of Biochemistry, College of Physicians and Surgeons, Columbia University, New York.* Groups of young rats were maintained on a synthetic, fat-free diet and were placed on a deuterium oxide regimen for 0.5, 1, 2, 4 and 8 days prior to being killed. The concentrations of deuterium were then determined in the glycogen of liver and of carcass, in the liver and depot fatty acids, in the cholesterol of liver and of carcass, in the liver protein, and in the body water. Three series of such experiments have been completed; first, a control series receiving no medication; second, a series in which the rats received thiouracil in their drinking water at a concentration of 0.1%; and, third, a series in which 1% of desiccated thyroid gland was added to the diet. From the rates at which the various tissue constituents became enriched with isotope, alterations in the rates of various anabolic processes produced by the medications employed could be inferred.

Effect of zinc-hydrochloric acid hydrolysis on the estrogens in human urine. BENJAMIN F. STIMMEL. *Rees-Stealy Medical Research Fund, San Diego, Calif.* Previous work in this laboratory has shown that there is some chemical confirmation for Smith and Smith's observation that the enhanced estrogenic activity of urine extracts prepared by zinc-hydrochloric acid hydrolysis is greater than can be accounted for by assuming conversion of estrone to α-estradiol and more complete hydrolysis. Further evidence has been derived, with our liquid chromatogram technique, from studies with human pregnancy urine and with non-pregnancy urine from estrogen treated subjects which would indicate that zinc-hydrochloric acid hydrolysis (1) converts the major portion of estrone to estradiol, (2) yields estradiol titers which are much higher than can be accounted for by conversion of estrone to estradiol, (3) yields estriol titers which appear higher than can be accounted for by more complete hydrolysis. Subsequent zinc-hydrochloric acid hydrolysis of residual urine after hydrochloric acid hydrolysis and ether extraction yields sufficient estrogen to account for a major portion, but not all, of the enhanced titer of zinc-hydrochloric acid hydrolyzed urine. Zinc hydrochloric acid hydrolysis of the phenolic residue from hydrochloric acid hydrolysis caused almost complete destruction of the estrogen present. These observations would appear to offer additional evidence in support of the theory that hydrolysis of the urinary estrogens in the presence of a reducing medium rehydrogenates certain estrogen metabolites. These hypothetical metabolites appear in the estradiol and estriol fractions of our liquid chromatogram.

Inhibition of pteroylglutamic acid conjugase by glutamic acid peptides of p-aminobenzoic acid E. L. R. STOKSTAD, J. PIERCE (by invitation), T. H. JUKES and A. L. FRANKLIN (by invitation). *Lederle Laboratories Division, American Cyanamid Company, Pearl River, New York.* The action of glutamic acid peptides of *p*-aminobenzoic acid on the activity of a conjugase preparation which converted conjugated forms of pteroylglutamic acid (PGA) to free PGA was studied. It was found that *p*-aminobenzoyl-gamma-glutamyl-gamma-glutamylglutamic acid at a concentration of .001 M reduced the activity of chicken pancreas conjugase by 80%. The tetraethyl ester of the same peptide produced no inhibition. P-nitrobenzoyl-gamma-glutamyl-gamma-glutamylglutamic acid was only about half as active an inhibitor of conjugase as the corresponding *p*-aminobenzoyl compound. The activity of simpler peptides was also investigated.

Interrelationships between pteroylglutamic acid, adenine, thymine, and antagonists of pteroylglutamic acid. E. L. R. STOKSTAD, M. REGAN (by invitation), A. L. FRANKLIN (by invitation), and T. H. JUKES. *Lederle Laboratories Division, American Cyanamid Company, Pearl River, New York.* It was previously reported (Franklin, A. L., Stokstad, E. L. R., Belt, M. and Jukes, T. H., J. Biol. Chem., **169,** 427 (1947)) that a crude "pteroylglutamic acid antagonist" showedco mpetitive inhibition against pteroylglutamic acid (PGA) when measured with *Streptococcus faecalis* R as a test organism. Pteroylaspartic acid was found to show competitive inhibition against PGA (Hutchings, B. L., Mowat, J. H., Oleson, J. J., Stokstad, E. L. R., Boothe, J. H., Waller, C. W., Angier, R. B., Semb, J., and SubbaRow, Y., J. Biol. Chem., **170,** 323 (1947)) and the growth-promoting effect of thymine was found not to be reversed by pteroylaspartic acid. In the present investigation a purified culture medium was used from which PGA, adenine and thymine were omitted. The crude antagonist was found to inhibit growth of *S. faecalis* R in the presence of the following combinations: PGA; PGA *plus* adenine; PGA *plus* thymine. However, when both thymine and adenine were added to the culture medium, growth was stimulated and the crude antagonist had no inhibitory effect. The effect of another PGA antagonist, N^{12}-methylpteroic acid (Cosulich, D. B., and Smith, J. M., Jr., J. Am. Chem. Soc., in the press), was

also studied in relation to purines and pyrimidines with *S. faecalis* R. Experiments were also carried out with rats and chicks. Thymine produced little or no growth or hematological response in rats maintained on a PGA-free diet containing succinylsulfathiazole, nor was thymine able to protect rats against the deficiency syndrome produced by the crude antagonist. Thymine at levels of from 0.5% to 2% in the diet was ineffective in protecting chicks against PGA deficiency on a purified diet with PGA omitted.

The role of acetylphosphate in the phosphoclastic and dismutation reactions of pyruvate. HAROLD STRECKER (by invitation) L. O. KRAMPITZ and HARLAND G. WOOD. *Department of Biochemistry Western Reserve University Cleveland 6, Ohio.* The occurrence of acetylphosphate as an intermediate in the phosphoclastic reaction ($CH_3 \cdot CO \cdot OOH + H_3PO_4 \rightleftharpoons CH_3CO \cdot OPO_3H_2 + HCOOH$) with *E. coli* is indicated by the formation of a labile phosphate compound and furthermore the reversibility of the reaction appears likely as judged by fixation of formate (Lipmann et al and Utter et al). Nevertheless, failure of numerous investigators to demonstrate activity of synthetic acetylphosphate with other enzyme preparations prompted further tests with *E. coli*. The following carbon labeled compounds, $C^{13}H_3 \cdot COOH$, $CH_3 \cdot C^{13}O \cdot OPO_3H_2$, $HC^{14}OOH$ were synthesized and added simultaneously to non-labeled pyruvate which was fermented to partial completion. With our enzyme preparation acetate, formate, lactate and CO_2 were products of the reaction indicating that part of the pyruvate under went a dismutation reaction ($2CH_3 \cdot CO \cdot COOH \rightarrow CH_3 \cdot CHOH \cdot COOH + CH_3 \cdot COOH + CO_2$). The residual pyruvate of the fermentation was degraded by yeast carboxylase with formation of CO_2 from the carboxyl group and acetaldehyde from the α and β carbons. The carboxyl carbon contained a high concentration of C^{14} but there was no C^{13} in the α and β carbons. Since carbon from $NaHC^{14}O_3$ was not fixed in significant amounts under the conditions of the experiment, the C^{14} of the formate apparently did not enter as CO_2. The results indicate that formate is fixed in pyruvate without occurrence of acetate or acetylphosphate as essential intermediates. It is concluded that either synthetic acetylphosphate is not an active component of the phosphoclastic reaction or there is another mechanism for the fixation of formate. It is considered possible that the chemically synthesized acetylphosphate is not identical with biological acetylphosphate.

A comparison of reactions of cysteine and beta-beta dimethyl cysteine. M. X. SULLIVAN. *Chemo-Medical Research Institute, Georgetown University, Washington, D. C.* Over a period of years data have been given to indicate that the Sullivan test for cysteine requires the presence of free -(SH), -(NH_2), COOH and in the arrangement in which these groups occur in cysteine. It was shown by Carter in 1941 that mono-methyl cysteine gave the characteristic color when Lugg's 1933 modification of the Sullivan test for cystine was used. Carter, however, found that one isomer gave the same amount of color as an equivalent amount of cysteine while the other isomer gave only 20% as much color. While in this laboratory S-methyl cysteine was found negative in the Sullivan cysteine reaction as should be expected no study was made as to the effect of substitution on the alpha or the beta carbon. Recently, however, we had occasion to study beta-beta di-methyl cysteine and found it absolutely negative in the cysteine reaction and also negative in the cystine procedure. However, in the cystine procedure it gives a positive reaction up to the addition of $Na_2S_2O_4$, that is it behaves like cystine amine. Data will be given as to the reason for the negative Sullivan reaction and for possibility of testing for the beta-beta di-methyl cysteine as an entity and as a decomposition product of penicillin.

Effect of pH on the aerobic metabolism of lymphosarcoma cells. WILLIAM H. SUMMERSON, HELENA GILDER (by invitation), and JOHANNA M. LEE (by invitation). *Department of Biochemistry, Cornell University Medical College, New York, N. Y.* The aerobic metabolism of the Gardner mouse lymphosarcoma cell has been studied *in vitro* in bicarbonate-containing media over the pH range 6.9 to 7.6. At pH 7.4, oxygen consumption averaged 0.80 microliters per 10^6 cells per hour, and R.Q. averaged 0.83; neither value showed any marked alteration with change in pH. By contrast, aerobic acid production proved to be dependent upon pH in a substantially linear fashion, with values ranging from 0.30 microliters of acid per 10^6 cells per hour at pH 6.9 1.05 microliters at pH 7.6. All of the acid produced aerobically by this tumor cell is lactic acid. Glucose utilization by the cell is substantially accounted for in terms of lactic acid produced. Hence aerobic glucose utilization is likewise a function of the pH of the medium, and the substrate for oxidative metabolism is not exogenous glucose. The bearing of these findings on current concepts of tumor metabolism will be discussed.

Influence of the concentration of the vitamin B complex on protein utilization. BARNETT SURE and FREELAND ROMANS (by invitation). *University of Arkansas, Fayetteville.* Seven concentrations of the vitamin B complex were used. These concentrates were fed to albino rats separately from the ration. The daily doses of thiamine, riboflavin, pyridoxine, and niacin varied from 3 to 100 μg. The doses of calcium pantothenate ranged from 25 to 600 μg; those of p-aminobenzoic acid, from 0.25 to 12 mg; inositol, from 0.075 to 3 mg; and choline chloride, from 3 to 12 mg. Three levels of purified casein

(Smaco) were used; as follows: 7.1% and 8.7%, supplemented with 25 mg cystine daily, and 17.3%; also, 9% lactalbumin (Borden). On the 7.1% and 8.7% levels of casein intake and on the 9% lactalbumin ration the optimum protein utilization was obtained on the concentration of the vitamin B complex, which furnished the following daily doses; 3μg thiamine, 3 μg pyridoxine, 3 μg niacin, 25 μg calcium pantothenate, 250 μg p-aminobenzoic acid, 75 μg inositol, and 3 mg choline chloride. On the 17.3% protein level, no notable differences were found until after the animals were ten weeks on such diet, when again the same low concentration of the B complex proved to be the optimum. The greatest transition occurred on the 7.1% casein level when changed from 3 to 5 μg daily doses of thiamine, riboflavin, pyridoxine, and niacin, and when the rest of the components of the vitamin B complex was doubled. The biological value of the casein was thus increased from 48.5 to 85.5.

A glycogenolytic factor from pancreas. Earl W. Sutherland and Christian de Duve (introduced by Carl F. Cori). *Dept of Biochemistry, Washington Univ. School of Med., St. Louis.* Liver glycogenolysis has been reported previously to be stimulated in vivo and in vitro by a non-dialyzable factor which is present in some insulin preparations and which, unlike insulin, withstands incubation with dilute alkali. Extracts of pancreatic tissue from dogs, rabbits and cattle, prepared by a method which is known to extract insulin, were found to increase glycogenolysis in liver slices. Maximal activity in the test system was obtained with amounts of extracts representing 0.05 to 0.20 grâms of pancreas; addition of more glycogenolytic factor did not further increase glycogenolysis. The activity was retained after dialysis and after alkali incubation. Extracts of spleen, liver and kidney were completely inactive. Slight activity was found in duodenal extracts. Extracts of fetal calf pancreas showed maximal activity in amounts equivalent to 0.02 to 0.04 grams. Pancreatic extracts from two alloxan diabetic rabbits showed essentially the same activity as those from normal rabbits and caused hyperglycemia without subsequent hypoglycemia when injected intravenously into a rabbit. Some material showing similar activity was found to be present in concentrated perfusates of isolated dog's pancreas and, in some cases, in alcoholic extracts of lyophilized dog's serum. Good recovery of the glycogenolytic factor was obtained when a small amount of an insulin preparation containing the factor was treated by these methods and no activity arose from similar treatment of an insulin preparation free of the factor.

An enzyme requiring DPN involved in the metabolism of testosterone by liver tissue. Max L. Sweat (by invitation) and L. T. Samuels. *Department of Biochemistry, University of Utah School of Medicine, Salt Lake City, Utah.* An active cell-free extract which will metabolize testosterone has been prepared from rat liver. The rate of the reaction is enhanced by the addition of diphosphopyridine nucleotide. It has been found that both malate and lactate inhibit the destruction of testosterone. The addition of nicotinamide increases the rate of destruction. The inhibition produced by these two DPN-requiring substrates, together with the increased rate of destruction in the presence of nicotinamide is strong evidence that the testosterone destroying enzyme is another enzyme which required DPN as a co-factor. The products of the reaction in which DPN is a coenzyme appear to be 17-ketosteroids without a conjugated double bond system. Work on the further purification of the enzyme is in progress.

A procedure for the determination of 4-aminosalicylic acid in blood and urine. David M. Tennent and Margaret L. Leland (introduced by Randolph T. Major). *Merck Institute for Therapeutic Research, Rahway, N. J.* A procedure has been developed for the determination of microgram amounts of 4-aminosalicylic acid in blood and urine. This depends upon two suitable color reactions which have been studied. The color produced is red when the drug is coupled in acidic solution with diazotized p-nitraniline followed by the addition of sodium hydroxide; and blue when the drug and p-nitraniline are both diazotized, followed by coupling in the presence of pyridine and then by addition of sodium hydroxide. The colors due to interferring substances in urine and in blood filtrates are equal in intensity with either procedure and their effect can be eliminated by taking two samples for the determination, developing the red color in one and the blue in the other, and reading in the colorimeter at 620 mμ, using the former as the blank for the latter. In practice the same reagents are used in both reactions, and the color developed depends upon the order in which they are added. Amounts of 4-aminosalicylic acid between 1 and 15 micrograms in 4 ml. of solution can be determined within ±5% by this procedure. It is applicable to blood filtrates prepared with tungstic acid and to urine dilutions.

Some factors which influence methionine excretion in the rat. Herbert C. Tidwell (introduced by Max N. Huffman). *Department of Biochemistry, Southwestern Medical College, Dallas, Texas.* The effect of changes in the protein and fat content of the diet, of fasting, and of the injection of methionine and choline on the urinary excretion of methionine has been investigated. Variations in the dietary protein during 4 or 33 days, with methionine intakes of 6 to 60 mg. per 100 gm. body weight, did not alter the methionine excreted. Unlike the results obtained by Homburger (Am. J. Med. Sci. **212**, 68, 1946) on normal men, similar

excretion values were obtained on all these animals after injecting supplements of the free amino acid equal to one or two times the normal daily intake. Less methionine was excreted after 15 or 40% fat diets than a 5% one. Still less was excreted after a 3 day fast. However, again a like amount of the injected supplement was excreted whether the animals were fasted, or on the high fat diets. The injection of choline during the 3-day fast was accompanied by an increased methionine excretion. The decreased excretion after higher fat intakes, or fasting, might result from an increased need of lipotropic substances associated with an increased fat metabolism. The excretion of a similar amount of the injected amino acid, whether a deficiency existed or not, suggests that increased needs are not reflected by a marked decrease in excretion of the supplement. The small increases in excretion following these supplements indicate that the amino acid is largely utilized, either as the amino acid or some degradation products.

On the bacterial metabolism of lysine. GERRIT TOENNIES and DOROTHY L. GALLANT (by invitation). *Lankenau Hospital Research Inst. and Inst. for Cancer Research, Philadelphia 30, Pa.* If in our experimental medium for the determination of amino acids with *S. fecalis* (*Fed. Proc.*, **6**, 298 (1947)) the concentration of lysine is suboptimal initial growth, which is proportional to the available lysine, is followed by rapid lysis of cells. An excess of 5 x of lysine per cc is sufficient to prevent lysis. In the lysine-limited medium lysis is prevented by omission of 98% of the phosphate buffer (0.3 *M*, pH 6.5), or its replacement by citrate buffer or sodium chloride of equal ionic strength. The irreversibility of the deamination of lysine (Schoenheimer) may explain the necessity of an excess of this particular amino acid for bacterial maintenance if phosphate accelerates amino acid exchange metabolism. However, lysis does not result from an interaction between cells and the lysine-deficient high-phosphate medium but expresses a property of cells grown in such a medium. Gale (*J. Gen. Microbiol.*, **1**, 53 (1947)) has demonstrated in another strain of *S. fecalis* that normal cells contain substantial amounts of free lysine while bacteria grown in a medium of low lysine content contain very little of the free amino acid. A similar deficiency in internal free lysine would explain the difference between our cells grown in a high-phosphate medium in the presence or absence of excess lysine. A medium containing only salts, lysine and glutamic acid prevented lysis, in accordance with Gale's finding that such a medium enables lysine-deficient cells to imbibe lysine. We also confirmed that glucose cannot replace glutanic acid.

The influence of KCN and pteroylglutamic acid on growth and prophyrin production of *Corynebacterium hoffmannii* JOHN R. TOTTER and EDITH S. SIMS (by invitation). *Department of Biochemistry, School of Medicine, University of Arkansas, Little Rock.* The organism *Corynebacterium hoffmannii* was grown in a basal medium complete for *Streptococcus faecalis* except for omission of pteroylglutamic acid (PGA). As supplement sterile KNC neutralized with acetic acid was added aseptically up to concentrations of 0.001, 0.002, and 0.003%. PGA was added to the medium before sterilizing in concentrations of 0.0, 0.005, 0.05, 0.1, and 0.2 mg. per cc. Porphyrins were extracted and determined by the customary procedures after 5–8 days incubation at 32°. Growth was determined by measuring optical density at suitable intervals. The KCN retarded the growth somewhat but did not completely inhibit growth even at 0.005% concentration. The porphyrin production was diminished by 7% in the 0.001% KCN, by 20% in the 0.002% KCN, and by 70% in the 0.003% KCN when measured on equal volumes of culture suspensions with the same optical density and compared with a control. PGA at concentrations above 0.05 mg. per cc. counteracted the effect of KCN on both growth and porphyrin production. Accelerated growth was also seen at relatively high PGA concentrations in the medium without KCN. The actual concentration at which PGA became effective varied considerably for reasons not yet ascertained.

The influence of fat in the diet upon nitrogen metabolism and liver protein regeneration. HARRY M. VARS and CHARLES E. FRIEDGOOD (by invitation). *Harrison Dept. of Surgical Research, Schools of Medicine, Univ. of Pennsylvania, Philadelphis.* In a previous study (Fed. Proc., 6, 257, 299), the diets used were made to contain only 3% of fat, 7% of the calories. From the work of Forbes, Deuel, and of others, the inclusion of adequate amounts of fat in the diet appears to improve the utilization of energy and the physical fitness of normal animals. Therefore, we have investigated the effect of adding 10% and 30% fat to a non-protein diet and to diets containing casein, upon liver protein regeneration and N metabolism after partial hepatectomy. Groups of rats were depleted of protein stores by a 2-week period of non-protein feeding, and 69% of their livers removed. They were then fed diets containing 10% and 30% fat for 2 weeks. Total N balances and new liver protein formed were determined. The data so obtained have been compared with the earlier observations. An increased percentage of fat calories in the non-protein diet did not increase conservation of N as measured by liver protein regeneration, although it decreased the negative N balance. Fat added to a casein diet in which the protein to fat calorie ratio remained constant, caused a greater food intake with an associated increase in liver protein regeneration and N balance. This was also true in

unoperated controls. Ingestion of an increased percentage of calories as fat during a period of isonitrogenous isocaloric feeding produced an increase in liver protein regeneration. This was accompanied by less retention of N by the body.

An electrophoretic analysis of the interaction of aldolase and glyceraldehyde-phosphate dehydrogenase with phosphate ions. SIDNEY F. VELICK. *From the Department of Biological Chemistry, Washington University School of Medicine.* Both aldolase and d-glyceraldehyde phosphate dehydrogenase contain an excess of basic over free acidic groups. If the individual groups were to exhibit dissociation contants in the range usually observed in other proteins one would predict alkaline isoelectric points whereas both proteins in phosphate buffers of ionic strength 0.1 are isoelectric on the acid side of neutrality. Since the substrates of both enzymes are anions one would attribute to them an affinity for anions if their kinetics were formulated in terms of an enzyme substrate complex. The acid isoelectric points can be explained by the binding of phosphate ions. Electrophoretic analysis shows that the isoelectric point of aldolase shifts from 6.7 to 4.94 in phosphate buffers ranging from ionic strength 0.05 to 0.2. The dehydrogenase shows a similar variation. The bound charge as a function of pH and ionic strength can be calculated and dissociation constants can be derived. Other properties of the dehydrogenase are consistent with its tendency to bind anions.

In vitro utilization of glucose by rat diaphragm muscle. C. A. VILLEE, F. M. SINEX, and A. K. SOLOMON (introduced by A. B. HASTINGS). *Department of Biological Chemistry and the Biophysical Laboratory Harvard Medical School.* The experiments of Gemmill, Stadie, and Krahl and Cori have demonstrated an effect of insulin on the formation of glycogen by isolated diaphragm muscle. Experiments were undertaken to study the oxidation of glucose by diaphragm muscle *in vitro* under varying experimental conditions. Glucose labeled with C^{14} in positions 3 and 4 was made biosynthetically as glycogen in rabbit liver slices from pyruvate and $C^{14}O_2$ by the Buchanan and Hastings method. Rat diaphragm muscle was incubated in Warburg vessels in a medium of 0.04 M sodium phosphate, 0.005 M $MgCl_2$, 0.08 M NaCl, and 0.2 per cent glucose, initial pH 6.8. A half diaphragm was placed in each vessel; to one of each pair of vessels, 0.5 units insulin per ml. medium was added. The CO_2 collected in the center well was precipitated as $BaCO_3$, plated and counted. Samples of the media before and after incubation were analyzed for glucose, and the glycogen in the diaphragm after incubation was hydrolyzed and determined as glucose.

In normal diaphragm muscle, from 50 to 80 per cent of the glucose which disappears from the medium is oxidized, the remainder is converted to glycogen. The addition of insulin increases both glucose oxidation and glycogen formation, although there is no over-all increase in respiration. The rates of glucose oxidation by muscle from alloxan-diabetic and from adrenalectomized rats are significantly lower and higher, respectively, than normal. These results support Cori's views that insulin affects primarily the hexokinase reaction.

Studies on the composition of nucleic acids. ERNST VISCHER (by invitation) and ERWIN CHARGAFF. *Department of Biochemistry, College of Physicians and Surgeons, Columbia University, New York.* The method for the micro partition chromatography of purines presented recently (E. Vischer and E. Chargaff, J. Biol. Chem., **168,** 781 (1947)) has been extended to the pyrimidines, cytosine, uracil, and thymine. It now permits not only the identification of the nitrogenous constituents of nucleic acids but also their quantitative estimation in minute amounts (5γ* of the individual component) with an accuracy of ± 4%. For the complete analysis of a nucleic acid 8 mg. were employed; but even this small sample weight can doubtless be reduced. A study of the acid hydrolysis of yeast ribonucleic acid showed that while for the estimation of purines the customary treatment with dilute acid could be employed, the pressure hydrolysis with 20% HCl for the quantitative liberation of pyrimidines was not applicable since it resulted in an extensive conversion of cytosine to uracil. Formic acid could, however, be used. Two different solvent systems served for the separation: a mixture of butanol, morpholine, diethylene glycol, water for the purines; butanol-water for the pyrimidines. Results obtained by the use of this technique will be presented for a representative series of nucleic acids, viz., the pentose nucleic acids of yeast and pancreas and the desoxypentose nucleic acids of yeast, tubercle bacilli, thymus, and spleen.

The inhibition of glutaminase by glutamic acid. HEINRICH WAELSCH and PHYLLIS OWADES (by invitation). *Depts. of Biochemistry, New York State Psychiatric Institute and Columbia University.* Glutamine appears not to participate in certain metabolic conversions in which glutamic acid is implicated. In the tissues studied the amide occurs in considerably higher concentrations than the parent amino acid and its formation and breakdown may be one of the regulating mechanisms of glutamic acid metabolism. The glutaminase of brain or kidney is inhibited by glutamic acid (Krebs). This inhibition is dependent on pH, being smallest in the range of optimal enzyme activity. At pH 6.8 (0.1 molar phosphate) 0.015 molar glutamic acid inhibits the splitting of 0.004 molar glutamine by rat brain homogenate by about 90

per cent and at pH 7.9 by about 15 per cent. It has been shown that the glutaminase of brain is active only in the presence of certain anions such as phosphate, sulfate, arsenate (Carter and Greenstein) or citrate. The extent of the inhibition of glutaminase by glutamic acid is dependent on the concentration of phosphate (or sulfate or citrate); also in the pH range of optimal enzyme activity a strong inhibition can be obtained at low phosphate (0.025 molar) concentrations. Variations in the pH or in the phosphate concentration may therefore determine the rate of enzymatic liberation of glutamic acid from glutamine.

Serine content of purothionin. M. K. WALDEN. (Introduced by A. K. Balls) *Enzyme Research Laboratory Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, USDA, Albany, California.* The protamine-like substance obtained from wheat flour as a crystalline hydrochloride by Balls, Hale and Harris (Cereal Chem., **19,** 279 (1942)) has been found to contain serine. The presence of serine supports the idea that in nature purothionin is linked with a phospholipid which is responsible for the solubility of the mother-substance in petroleum ether and the loss of that solubility, as well as of associated phosphorus, on hydrolysis to purothionin.

After hydrolysis of the recrystallized hydrochloride with constant boiling HCl for 20 hours, formaldehyde equivalent to 9.8 percent of serine (by weight) was obtained by the method of Boyd and Logan (Jour. Biol. Chem., **146,** 279 (1942)). A phosphotungstic acid separation after Van Slyke, Hiller and MacFadyen (Jour. Biol. Chem., **141,** 681 (1941)) excluded the possibility of hydroxylysine; 91 percent of the expected serine was found in the filtrate. No ethanolamine was found by the method of Blix (Biochem. Zeits., **305,** 129 (1940)) and the residue remaining gave formaldehyde equivalent to 9.5 percent serine in the original substance. No formaldehyde was produced by periodate from the unhydrolyzed material. Arginine, serine and cystine thus account for more than half of the total nitrogen of the substance, and occur, respectively, in proportions of equivalents close to 2:2:1.

Inhibitors of choline acetylase. MICHAEL S. WEISS, HELEN VORIPAIEFF, and DAVID NACHMANSOHN. (Introduced by H. T. CLARKE). *Dept. of Neurology, College of Physicians and Surgeons, Columbia University, New York.* The accelerating or inhibitory effects of some intermediate compounds on enzymes depends frequently on the degree of purity. Apparent contradictory observations of this type of effects are often due to the use of different enzyme preparations. Such effects may be the result of the reactions with other intermediate metabolic processes and not with the enzyme system itself.—Citric and L glutamic acid increase the rate of freshly prepared and dialyzed choline acetylase. No effect is observed with glutamic acid in active enzyme solutions prepared from acetone-dried powder in presence of active coenzyme. The effect of citric acid seems to change with purification. In highly active solutions prepared from the head ganglion of Squid in which one gram of protein formed 150 milligrams of acetylcholine per hour, citric acid decreased the rate of formation in 10^{-2}M concentration by about 50 per cent. The effect may be due to the removal of magnesium necessary for transphosphorlyation. α-keto glutaric acid which like other α-keto acids inhibits all preparations obtained so far from mammalian brain, had no effect on the Squid ganglion preparation.

Among the inhibitors of choline acetylase which are not naturally occurring compounds, the naphthoquinones are of particular interest. A considerable number of them, for which we are greatly indebted to Professor Louis F. Fieser, were tested and some of these effects will be described. The 2-Methyl-1,4-naphthoquinone-8-sulfonic acid (K salt) was found to have the same effect whether tested on the mammalian brain or Squid ganglion preparation, suggesting in this case an affinity to the enzyme system itself.

Ozonolysis of the pyo compounds. IBERT C. WELLS (by invitation), WILLIAM H. ELLIOTT (by invitation), SIDNEY A. THAYER and EDWARD A. DOISY. *Laboratory of Biological Chemistry, Saint Louis University School of Medicine, Saint Louis, Missouri.* As a continuation of our work on the structure of the antibiotic substances produced by *Pseudomonas aeruginosa* (J. Biol. Chem., **159,** 725 (1945)), some of them have been subjected to ozonolysis. Pyo III ($C_{34}H_{44}N_2O_2$) yielded one mole of n-octanal (isolated as the 2,4-dinitrophenyl hydrazone, m.p. 106–107°C). No other products were isolated. Pyo Ic (tetrahydro Pyo III) treated in the same manner yielded no aldehyde or ketonic fragments but did produce N-caprylanthranilic acid, capric acid and capramide. These compounds were identified by ultimate analyses and comparison with appropriate derivatives. Anthranilic acid was detected spectroscopically but was not isolated. The yield of N-capryl anthranilic acid and the estimated yield of anthranilic acid accounted for one mole of N-capryl anthranilic acid per mole of Pyo Ic. Octahydro Pyo Ic (dodecahydro Pyo III) produced upon ozonization one mole of capramide and a small amount of a new compound which is acidic, melts at 131–132°C., does not absorb ultra-violet light and analysis indicates the formula $C_{17}H_{35}NO_4$.

Acetaldehyde metabolism in dogs maintained on a purified diet. W. W. WESTERFELD and J. M. MCKIBBIN (by invitation). *From the Department of Biochemistry, Syracuse University College of Medicine, Syracuse, New York.* Young adult dogs

were maintained on a purified diet containing casein 19, cod liver oil 2, cottonseed oil 7, salts 4, sucrose 28, glucose 40, and adequate amounts of thiamine, riboflavine, pyridoxine, pantothenate, choline, nicotinic acid and tocopherols. Within 4 months approximately 50% of the dogs developed a defect in acetaldehyde metabolism that was brought out by a load test in which 80 mg AcH per kg were administered intravenously and its rate of disappearance from the blood was measured under standardized conditions. A delayed disappearance curve was not associated with weight or skin changes, but was associated with a liver pathology histologically similar to hydropic degeneration and identical with that attributed by Elman and Heifetz (J. Exp. Med., **73**, 417, 1941) to protein deficiency. Other evidence of liver damage was not prominent; liver fat was normal; brom sulfalein excretion and prothrombin times were normal; serum alkaline phosphatase may have been slightly elevated.

Once this defect was produced, it was difficult to restore the acetaldehyde disappearance curve to normal by dietary means. The only pure vitamin that gave a positive response was inositol, and this effect was very erratic; it produced a beneficial response in only some of the dogs, and the response was often temporary. Negative tests were obtained with biotin, folic acid, p-aminobenzoic acid, vitamins C, K, and E, and with various combinations of these factors. Disappearance curves in some of the dogs could not be restored to normal even by feeding natural foodstuffs, such as meat, liver, milk, yeast, or molasses.

The effect of streptomycin on deamination and oxygen consumption by resting cells of E. coli. KENT WIGHT (by invitation) and DEAN BURK. *National Cancer Institute, National Institute of Health, Bethesda, Maryland.* Warburg technique studies were carried out on the mode of action of streptomycin on resting cells of *E. coli*, strain NIH 119, grown 16 to 24 hours in nutrient broth at 37°C, centrifuged, washed and resuspended in 0.05 M phosphate at pH 7.4, and then studied 4 to 5 hours at 37°C, with and without addition of 10 to 20 p.p.m. streptomycin, 0.012 M amino acid, or 0.007 M non-nitrogenous organic acid.

Anaerobically, in N_2, the washed, resting *E. coli* cells deaminated aspartic acid and serine, but not threonine, alanine, or histidine. Streptomycin inhibited this ammonia production up to 50% in the case of aspartate and 35% with serine. No CO_2 production was detected with any of the five amino acids, even in the presence of 0.01–0.02 M semicarbazide. Oxaloacetate and pyruvate, but not acetate, yielded CO_2, without inhibition by streptomycin, however.

In air, ammonia production from aspartate and serine was somewhat greater than in N_2, and the percentage inhibition by streptomycin was somewhat less (20–30%). At the same time, oxygen consumption (Q_{O_2} = ca. 30) was decreased about 50% by the fourth hour by streptomycin. Threonine and alanine gave the same oxygen consumption as aspartate, but less NH_3 production, and no inhibition of either process by streptomycin. The Q_{O_2} with fumarate was decreased by streptomycin from 40 to 25, and with oxaloacetate from 90 to 50. However, streptomycin did not inhibit either the aerobic or anaerobic assimilation of ammonia when added with fumarate, the presumed end-product of aspartate deamination.

Variables affecting the assay of testosterone propionate using male castrate rats. G. A. WILLS, SYBIL RAMPTON (by invitation) and L. I. PUGSLEY. *Food and Drugs Division, Dept. National Health and Welfare, Ottawa, Canada.* A study was made of the method of administration and of the time after castration on the precision and sensitivity of the method of assay of testosterone propionate described by Matheson and Hays (Endocrinology **37**, 275, 1945). It was found that the standard error of the assay and the dose required to give comparative responses was considerably decreased by using two injections intramuscularly instead of the one injection subcutaneously recommended by Matheson and Hays. It was confirmed that, providing two weeks were allowed for the regression of the seminal vesicles, the response was independent of the time after castration.

The effect of biotin on mitotic activity in the mouse liver. J. WALTER WILSON and ELIZABETH H. LEDUC (introduced by P. H. MITCHELL). *Brown University.* Biotin deficiency was produced in mice by inclusion of 20% dried egg white in a semi-synthetic diet. Five-week mice placed on this diet showed signs of deficiency in about 5 weeks; 16-day mice in 3–4 weeks. During the development of the deficiency the mice grew rapidly and in the younger ones mitotic activity was observed in the liver. Mice with well developed symptoms showed practically no mitosis in the liver. Deficient mice kept on the diet were treated with a single injection of homologous liver and sacrificed daily. Mitotic activity in their livers was observed on the 4th and 5th days (mitotic rates 1.41 and .95). In others injected daily with $1\frac{1}{2}\gamma$ of biotin intraperitoneally, or 2 γ of biotin subcutaneously, mitotic activity was found in the 1st and 2nd days (mitotic rates .21 and .79). In still others transferred to the same diet with the egg white replaced by casein, mitotic activity was observed from the 2nd to the 6th day with the highest rate on the 4th (mitotic rate 6.5). Non-deficient adult (15-week) mice were injected daily subcutaneously with 2 γ of biotin and one sacrificed each day. Mitosis was observed on the 3rd day (mitotic rate .95). There is thus a falling off of mitotic activity in the liver with actual development of symptoms of biotin deficiency, and a significant increase after treat-

ment of deficient animals with liver or biotin or simply after injection of biotin into normal adults.

Incorporation of C^{14} labeled glycine into the protein of tissue homogenates. THEODORE WINNICK, FELIX FRIEDBERG (by invitation), and DAVID M. GREENBERG. *Division of Biochemistry, University of California Medical School, Berkeley.* The incorporation of radioactive glycine into proteins was observed when homogenates of such tissues as rat spleen, liver, and kidney were incubated at 37° and pH 7.6 in the presence of the C^{14}-labeled amino acid. The process requires the presence of inorganic salts, oxygen, and certain organic metabolites, such as glucose and citrate. It is inhibited by azide or cyanide, and abolished by heating to 100°. In the case of spleen homogenates, the quantity of labeled glycine taken up per mg. protein was found to be proportional to both the glycine concentration in the medium and the reaction time. The activity of the homogenates was apparently associated with insoluble cell particles, which could be collected by centrifugation. Evidence that the process of glycine uptake represents an actual incorporation into the protein structure was provided by hydrolytic experiments. Following hydrolysis of the radioactive proteins with either proteolytic enzymes or hydrochloric acid, the C^{14} could be accounted for as glycine. The radioactivity of this glycine (recovered with the aid of carrier) corresponded fairly well to the C^{14} content of the initial proteins. It was estimated that approximately 0.2 per cent of the glycine residues of spleen proteins were replaced per hour by the labeled glycine added to the homogenate.

Thyroxine activity and antagonism of some structural analogues of thyroxine. RICHARD J. WINZLER and EARL FRIEDEN (by invitation). *Department of Biochemistry and Nutrition, the University of Southern California School of Medicine, Los Angeles.* A number of compounds structurally related to thyroxine have been prepared. These have included the dl-glycine homologue of thyroxine (3/5-diido-4-(3′,5′-diido-4′-hydroxyphenoxy) phenyl glycine) and the benzoic acid analogue of thyroxine (3/5-diido-4-(3′,5′-diido-4′-hydroxyphenoxy)benzoic acid) in which the side chain of thyroxine has been modified, and also dl-O-benzyl-n-acetyl-3,5-diiodotyrosine in which the prime ring has been altered. These compounds have been tested both for thyroactivity and for thyroxine antagonism using their effect on tadpole metamorphosis, their effect on basal metabolic rates in rats, and their influence on the weight of of thyroid glands of thiouracil-fed rats. The results indicate that the glycine homologue of thyroxine possesses about 10% of the activity of dl-thyroxine when measured with tadpoles but has been inactive in the rat at levels up to 1.5 mg./kilo/day. No thyroxine antagonism has been evident in studies with this compound. The benzoic acid analogue of thyroxine has been found to show about 20% of the activity of dl-thyroxine in tadpoles, but has been inactive in rats up to levels of 5 mg./kilo/day. The observations of Woolley (1) on the antagonistic action of O-benzyl-n-acetyl diiodotyrosine toward thyroxine in tadpole metamorphosis have been confirmed and extended. However, levels up to 50 mg./kilo/day showed neither thyroxine activity nor antagonism in rats.

A spectrophotometric method for nicotine in blood. WILLIAM A. WOLFF and MARINA HAWKINS (by invitation). *Tobacco Research Laby., Bowman Gray School of Medicine, Wake Forest College, Winston-Salem, N. C.* A colorimetric procedure for nicotine has been adapted for use with human blood and modified to increase its sensitivity and to eliminate the "nicotine blank" inherent in certain other methods. Blood proteins are precipitated with trichloroacetic or metaphosphoric acids and the filtrate is divided into aliquots, each equivalent to 10 ml. blood. One aliquot of filtrate is saturated with NaCl, made 20% with NaOH, and steam distilled in an all glass apparatus. The color value (nicotine plus "blank material") of the distillate is determined by the cyanogen bromide—B-naphthylamine reaction. The resulting color is compared with that of a nicotine standard similarly treated, both read at 490 millimicrons in a Beckman spectrophotometer. A second aliquot of acid filtrate is treated with an activated carbon which removes the nicotine and leaves the "blank material" in solution. The treated filtrate is distilled as above and its color value determined. The nicotine content of blood is the difference between the two color values. A series of control experiments with nicotine added to blood gave results between 90 and 100% of the amounts added. The sensitivity permits the estimation of 1 microgram nicotine in 20 ml. blood. This method is suitable for the study of nicotine levels in the blood of smokers. The nature of the "blank material" has not been determined. Its magnitude is not influenced by nicotinic acid or trigonelline.

Some crystalline peptides isolated from enzymic digests of insulin and their relationship to strepogenin. D. W. WOOLLEY. *The Laboratories of The Rockefeller Institute for Medical Research.* Because the amino group of strepogenin constitutes some of the free amino groups of insulin, the conversion of the protein to dinitrophenyl (DNP)-insulin, and subsequent digestion with proteolytic enzymes, should yield the growth factor in the form of DNP-strepogenin. This latter is colored yellow, and rendered distinct from most other products of digestion in that it is soluble in organic solvents. It may be differentiated from yellow impurities by the occurrence in it of glutamic acid.

Pancreatin digests of DNP-insulin, and of the DNP derivative of that portion of oxidized insulin which is soluble in water at pH 6, have been frac-

tionated by a series of countercurrent distributions between pairs of immiscible solvents, and several crystalline DNP peptides have been isolated. Aside from the yellow end group of DNP-glycine contained in them they yielded the following amino acids when hydrolyzed: (1) glutamic acid; (2) glutamic acid, serine, threonine, valine and leucine; (3) same as (2) plus aspartic acid; (4) glutamic acid, serine, tyrosine, and leucine; (5) same as (3) plus isoleucine and cysteic acid (from cystine). The regular increase in complexity of these substances suggested that the smaller compounds were split products of the larger. Their composition affords an insight into the structure of a portion of the insulin molecule. Furthermore, peptides (2) and (3) were isolated from digests of DNP-trypsinogen, from which fact it may be concluded that a sizable piece of the two proteins is identical. Finally, these observations may be of aid in the elucidation of the structure of strepogenin.

The application of paper chromatography to the study of amino-aciduria in patients with liver disease. N. F. Young and F. Homburger (Introduced by T. F. Gallagher). The method of paper chromatography developed by Martin and associates has made possible the separation and visualization of amino acids in complex mixtures. In this study this technique has been applied to the urines of patients with liver disease. Both hydrolyzed and unhydrolyzed urines from patients with infectious jaundice, homologous serum jaundice and hepatolenticular degeneration were chromatographed. The patterns so obtained made possible the estimation of the relative amount of each amino acid present. Total α-amino acid nitrogen excretion was determined on each sample by the ninhydrin reaction. The increase in α-amino acid nitrogen on hydrolysis is reflected in the chromatograms by increased amounts of some amino acids and the appearance of amino acids not found in unhydrolyzed urines. The results obtained will be compared to those obtained on normal urine. Although the degree and pattern of amino-aciduria were found to vary greatly they did not closely correspond to the clinical changes observed. Neither was the severity of the liver disorder as measured by standard liver function tests found to parallel the extensity of amino-aciduria. The significance of the results will be discussed with respect to dietary and renal factors as well as liver function.

AMERICAN SOCIETY FOR PHARMACOLOGY AND EXPERIMENTAL THERAPEUTICS, INC.

Thirty-Eighth Annual Meeting

Atlantic City, New Jersey, March 15, 16, 17, 18, 19, 1948

(For possible corrections in any of the following abstracts see the next issue)

Effects of amphetamine, dihydroergotamine, and methadon on human cerebral blood flow and oxygen uptake. Benedict E. Abreu and (by invitation) Grant W. Liddle, Arthur L. Burks, Alexander Simon, Violette Sutherland and Gilbert S. Gordan. *Divisions of Pharmacology and Experimental Therapeutics, and Psychiatry, University of California Medical School, San Francisco.* Cerebral blood flow and cerebral oxygen consumption were determined by the Kety-Schmidt nitrous oxide technic on a series of unanesthetized humans before and after administration of amphetamine, dihydroergotamine and methadon. Following the intravenous administration of amphetamine sulfate 20 mgm., mean arterial pressures increased approximately 30%, and cerebral blood flow and cerebral oxygen consumption were decreased by approximately 20% and 15% respectively. Dihydroergotamine 1 mgm., intramuscularly, had no significant overall effect on arterial pressure, cerebral blood flow or oxygen utilization. Amidone 5 mgm., intramuscularly did not produce conclusive effects.

Subject	Blood flow (ml./100 gm. brain min.)		Oxygen consumption (ml. O_2/100 gm. brain/min.)	
	Control	After drug	Control	After drug
Amphetamine sulfate				
1	70	54	4.3	3.6
2	36	31	2.7	2.3
3	70	58	4.1	3.5
Dihydroergotamine				
4	44	42	3.2	3.2
5	67	62	3.4	3.7
6	70	68	3.5	3.7
Methadon HCl				
7	57	70	3.2	4.1
8	41	38	3.0	2.4
9	71	69	4.2	4.3

Comparative effects of sympathomimetic amines on the vasomotor resistance of the kidney, mesentery and leg. RAYMOND P. AHLQUIST. *Department of Pharmacology, University of Georgia School of Medicine, Augusta.* Five sympathomimetic amines, closely related structurally to epinephrine, have been compared as to their effects on the vasomotor resistance (VR) of the kidney, mesentery and hind leg of the anesthetized, heparinized dog. VR equals the arterial pressure (P), in mm. Hg, minus 20, divided by the arterial flow (F), in cc. per min. P minus 20 was used since the relationship between pressure and flow is markedly altered when P is lower than 20 mm. Hg, due in part to the presence of the cellular elements in the blood. P was recorded by a Hamilton manometer, and F by a Shipley optical recording rotameter. Equimolar concentrations of the following racemic hydrochlorides were injected intra-arterially: I. arterenol, II. cobefrine, III. epinephrine, IV. 3,4-dihydroxy ephedrine, and, V. N-isopropyl arterenol. The results indicate that the vascular beds differ greatly in their sensitivity to sympathomimetic constrictors and dilators. In the kidney, III increases the VR the most; and IV and V have practically no effect. In the leg, however, I increases VR the most; and IV and V decrease the VR markedly. II is always less active than I in increasing the VR. The mesentery exhibits intermediate responses. The results also show that while epinephrine is the most active vasoconstrictor it is not the most active pressor agent, because it also possesses marked dilator activity which tends to diminish its pressor effect.

The effect of rutin on oxygen toxicity in rats. SHANNON C. ALLEN and TALBOT G. MORTAROTTI (by invitation). *From the Pharmacology Division, Bureau of Agricultural and Industrial Chemistry, U. S. Department of Agriculture, Albany, California.* It has long been known that high O_2 tensions are toxic and they have been shown by other investigators to inhibit certain of the enzyme systems involved in cellular metabolism. On the theory that such inhibition might be reversed by a physiologic antioxidant Puig Muset and Valdecasas (1946) exposed rats to 100% O_2 after pretreatment with vitamin P (eriodictyol) and found that it reduced the toxic effects. We have attempted to repeat their experiments using rutin and quercetin as vitamin P substances. Rutin, given in a single dose just before the experiment, had no effect on survival rate, weight loss or gross pathology of rats. When given in the diet and/or drinking water available throughout exposure to O_2, or in daily subcutaneous injections for 10 days previous to exposure, rutin and quercetin caused earlier deaths and fewer survivals. The most striking characteristic of the gross pathology of rats dying during exposure to O_2 is the marked hydrothorax and resulting lung collapse. There is a considerable body of experimental support for the theory that rutin exerts its physiological effects through the protection of circulating epinephrine and it has been shown by others that epinephrine has a dilator effect on pulmonary capillaries and enhances the susceptibility of young rats to O_2 poisoning. This effect of rutin is further substantiated by the observations of R. H. Wilson of this laboratory that rats treated with rutin are more susceptible than controls to death from pleural edema following thiourea administration.

Mode of action of antihistaminics agents. LILLIAN ALONSO (by invitation), MAXINE ADAMS (by invitation), LOUISE GODDARD (by invitation), MARION JAEGER (by invitation), and J. T. LITCHFIELD, JR. *Chemotherapy Division, Stamford Research Laboratories, American Cyanamid Company, Stamford, Connecticut.* Four antihistaminic agents, Pyribenzamine, Neoantergan, Chlorothen, and Benadryl, were studied for mode of action on atropinized guinea pig ileum. The concentrations of the antagonists used in these experiments ranged from 0.0004 micrograms/100 ml. to 40 micrograms/100 ml. of bath fluid, and those of histamine diphosphate from 0.3 micrograms/100 ml. to 5250 micrograms/100 ml. of bath fluid. For each antagonist the complete experiment was obtained on the same muscle strip, and a total of seventeen different experiments were carried out using the four different antagonists. The results of these studies indicate that the law of mass action is obeyed over a thousand-fold concentration range of both histamine and antagonist. These results are in agreement with the theory that histamine and the above antagonists compete for the same cellular receptors.

The effect of rutin and quercetin on scorbutic guinea pigs. ANTHONY M. AMBROSE. *From the Pharmacology Division, Bureau of Agricultural and Industrial Chemistry, U. S. Department of Agriculture, Albany 6, California.* Studies suggesting the use of flavone glycosides in the treatment of hemorrhagic manifestations of scurvy have been summarized (Nutritional Reviews 1943 and 1944). Evidence is based upon observations that foods rich in natural vitamin C are more effective than equivalent amounts of synthetic ascorbic acid in maintaining the integrity of the capillary wall. Studies have been undertaken to demonstrate the effect of rutin and quercetin in supplementing the action of subminimal doses of ascorbic acid in guinea pigs on a scorbutogenic diet. Young guinea pigs were placed on a scorbutogenic diet (Arch. Biochem., **12**: 375, 1947), to which was added Brewer's yeast to make 1%. Supplements of cod liver oil, 1 ml., were fed per os every 5 days. Other supplements, rutin 100 mgm., quercetin 100 mgm., (contained in ¼ ml. of propylene glycol), or as-

corbic acid 0.2 or 0.4 mgm. (contained in ½ ml. of water) were fed daily during the course of the experiment. Guinea pigs on the scorbutogenic diet when fed rutin or quercetin alone showed no improvement. However, when these same supplements were fed in conjunction with 0.2 or 0.4 mgm. ascorbic acid mortality, compared with ascorbic acid controls, was reduced and the general appearance of the animals improved. Tentatively it may be concluded that rutin or quercetin both appear to have a sparing action on subminimal doses of ascorbic acid in the scorbutic guinea pig. Quercetin appears to be more effective than rutin, but this may be explained by the difference in molecular weight of the two substances.

The effect of rutin on blood pressure in dogs and rabbits. ANTHONY M. AMBROSE. *From the Pharmacology Division, Bureau of Agricultural and Industrial Chemistry, U. S. Department of Agriculture, Albany 6, California.* The effect of rutin on blood pressure in experimental animals has been variously reported. Fukuda (Arch. exp. Path. pharmakol., **164:** 685, 1932) has reported a pressor effect in rabbits and frogs, while Czimmer (ibid. **183:** 587, 1936) has observed no effect on blood pressure in cats. In conjunction with studies on capillary permeability the effect of rutin on blood pressure has been studied in dogs, under intravenous barbital anesthesia, and in rabbits, under oral urethane anesthesia. In 5 dogs in which rutin was administered intravenously in doses of 5, 20, or 100 mgm./kgm. the effect on blood pressure was invariably depressor. The fall in blood pressure was in the order of 70 mm. Hg and usually returned to normal within 3 minutes. The size of the dose of rutin had no particular influence on the magnitude of the fall in pressure, but the time of restoration to normal pressure increased with the dosage. In 6 rabbits, in which 100 mg./kgm. was injected intravenously, five showed a fall in blood pressure similar to that observed in dogs, in two of which the blood pressure returned to within 10 mm. of the normal value and in three the blood pressure rose to about 20 mm. above the normal after the initial fall, with a gradual return to normal. In one rabbit the effect was initially pressor, followed by a depressor effect, and return to approximately normal within ½ hr. after administration.

Parasiticidal activity of thioarsenites in man. HAMILTON H. ANDERSON, HERBERT G. JOHNSTONE (by invitation), and A. PEÑA CHAVARRÍA (by invitation). *University of California Medical School, San Francisco, California, and Hospital San Juan de Dios, San Jose de Costa Rica, A. C.* The dithiocarboxymethyl and dithiocarboxyphenyl derivatives of p-carbamidophenyl arsenous oxide have exhibited activity against *E. histolytica in vitro* and in infected macaques (J. Pharmacol., **91:** 112, 1947). During the course of field trials in human amebiasis, the thioarsenites were employed against other human parasites. The adult (65 kilo) dose was 200 mgm. given orally thrice daily for 10 days. There were 12 persons harboring *Strongyloides stercoralis*, only one of whom was cleared; 2 with *Balantidium coli*, both cleared; 1 with *Fasciola hepatica*, with uncertain results; and 2 with *Leishmania tropica* and 3 with *Treponema pertenue*, none of whom were cleared. In each case tests of urine, blood, liver and kidney function showed no change during or after treatment, except increases in hemoglobin levels and body weight due to improved nutrition and bed rest. No evidences of drug intolerance developed except nausea in 2 which was controlled by salol coating of tablets.

The action of methadon upon the respiration of rat diaphragm and liver and kidney cortex slices. ARTHUR L. BACHELOR and HENRY W. ELLIOTT (introduced by BENEDICT E. ABREU). *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco.* The action of methadon upon the respiration of rat cerebral cortex slices has been previously reported (Elliott, Warrens, and James; J. Pharmacol., **91:** 98, 1947). The studies have been extended to include other tissues using methods described in the reference cited. Pieces of young rat diaphragm and liver and kidney cortex slices were subjected to concentrations of methadon which (a) stimulated (0.0005M), (b) produced a delayed inhibition (0.001M) and (c) strongly inhibited (0.002M) brain slice respiration. The results are summarized in the following table.

Tissue	Concentration					
	0.0005M		0.001M		0.002M	
	E	T	E	T	E	T
	%	*min.*	%	*min.*	%	*min.*
Diaphragm	153 110	30 90	120 57	15 90	134 5	15 90
Kidney	82	90	34	90	16	90
Liver	56	90	22	90	7	90

E = Q_{O_2} as per cent of control. T = Time of maximum effect.

The dual effect of methadon found for brain slices was shown to an even greater degree by diaphragm. Both 0.001 and 0.002M methadon produced a delayed inhibition of diaphragm respiration but all concentrations increased the oxygen consumption of diaphragm more than they stimulated brain respiration. There was no evidence of stimulation of liver or kidney cortex respiration at any time after addition of the drug. The selective stimula-

tion of brain and diaphragm respiration is in marked contrast to the action of dinitrophenol which has been shown to increase the oxygen consumption of all tissues included in these studies.

The maintenance of mean pulmonary arterial pressure after extreme right ventricular damage in the dog. AUGUSTUS C. P. BAKOS (introduced by CHARLES F. MORGAN). *Dept. of Physiology and Biophysics, Georgetown University School of Medicine, Washington, D. C.* Severe right ventricular damage was produced in barbitalized and heparinized dogs by repeated charring with a soldering iron as described by I. Starr (Am. Heart J. **26:** 291, 1943). Simultaneous mercury manometric recordings of mean systemic and pulmonary arterial pressure fluctuations were made from direct cannulations of the carotid and a right pulmonary arterial stem branch respectively. Peripheral venous pressure was determined directly by cannulation of the femoral vein. The degree of right ventricular damage produced was ascertained at the termination of each experiment by grossly estimating the area of epicardial surface charred and the depth of charring; thus over 75 per cent necrosis of the right ventricular muscle could be obtained. No gross pulmonary edema or congestion was noted at this time. The results of these preliminary studies on nine dogs showed that following the production of severe right ventricular damage the mean pulmonary and systemic arterial pressures had not changed significantly from pressures recorded prior to the damage. Similarly the peripheral venous blood pressure also remained fairly constant as previously reported by Starr. It was further observed that coincident with left ventricular systole the right ventricle showed passive enlargement and failure of active contraction. In two of these experiments, subsequently produced left ventricular damage resulted in a simultaneous decline in the pressures of both the pulmonary and systemic arterial systems. The relationship of these experiments to the dynamics of right ventricular failure is yet to be considered.

The adrenergic properties of antihistaminic drugs determined by electrical measurements. T. C. BARNES. *Dept. of Pharmacology, Hahnemann Medical College and Hospital of Philadelphia.* In the "oil-cell" method (Barnes and Beutner, Science **104:** 569, 1946) drugs are placed at the interface between saline and an oil layer and the resulting phase-boundary potential is measured. For example the negative potential produced by acetylcholine and prostigmine (Barnes, Arch. internat. Pharmacodynamie **73:** 386, 1947) may explain their stimulating action. The oil (representing the lipoid layer on the living cell) is held in a cup on a U-shaped glass tube immersed in saline. To avoid possible short circuits along the glass surface the drug solution and the saline on the opposite side of the oil layer may be introduced by two separating funnels dipping into a vessel of oil. The triglyceride oils have the special property of dissolving sympathomimetic drugs and thereby establishing a negative phase boundary potential (Barnes, Anat. Rec. **96:** 87, 1946, Seventeenth International Physiological Congress **60:** 1947). Acetylcholine has no effect on these oils. It was surprising to find that benadryl and pyribenzamine behave like sympathomimetic drugs on the triglyceride oils. 0.05% benadryl produced 40 mv. negative on triacetin in saline. 0.05% pyribenzamine produced 45 mv. negative on triacetin. Histamine has no electrical effect on triacetin. The results emphasize the possible adrenergic properties of these drugs. According to Sherrod pyribenzamine potentiates the pressor action of epinephrine. Loew has reported experimental bronchodilatation with benadryl.

Phase-boundary potentials of some analogs of prostigmine. T. C. BARNES. *Dept. of Pharmacology, Hahnemann Medical College and Hospital of Philadelphia.* Prostigmine has outstanding electrical activity *in vitro* (Barnes, Anatomical Record **96:** 87, 1946, Fed. Proc. **6:** 74, 1947. Arch. internat. Pharmacodynamie **73:** 386, 1947). The phase-boundary potentials of several analogs of prostigmine are shown in the table. Drugs were added to 200 cc. of aqueous medium containing the oil cup (Barnes and Beutner, Science **104:** 569, 1946). In the first column are numbers of the compounds. For chemical and biological data see Aeschlimann and Stempel in the Barell Volume, Basle, p. 306, 1946. The negative phase-boundary potentials in millivolts are recorded in the next four columns. Columns two and three are potentials produced by 0.001 molar concentrations in saline in contact with cresol and with guaiacol. The last two columns list potentials of one milligram and of ten milligrams at the interface between nitrobenzene and 200 cc. of equal parts saline and 0.1% sodium oleate. The data indicate the pronounced electrogenic action of all prostigmine analogs. Further studies may explain individual differences.

Prep.	0.001M on cresol	0.001M on guaiacol	1 mg. on oleate-nitrobenz.	10 mg. on oleate-nitrobenz.
	mv.	*mv.*	*mv.*	*mv.*
A. Ch.	44	28	75	110
Prostig	54	60	96	130
Nu-613	50	60	90	145
Nu-1250	40	57	90	155
Nu-1197	45	60	41	100
Nu-1208	40	60	47	122
Nu-1243	34	60	71	123
Nu-1249	47	60	69	130
Nu-1173	36	51	60	120

The toxicity of polyallyl alcohol. T. R. W.

BARNES (introduced by CHARLES H. HINE). *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco.* When administered intragastrically as a 50% aqueous solution, polyallyl alcohol produced no gross toxic effect in mice or rats which received as much as 27 gm./kg. Single administrations were made to 30 animals of each species, which were observed over 10 days. Skin irritation was tested on rabbits according to the method of Draize, et al. (J. Pharmacol., **82:** 377, 1944). A saturated solution of polyallyl alcohol in ethanol was applied to the shaved dorsal surface of albino rabbits. It produced neither irritation nor systemic effects in amounts up to 4.0 gm./kg. Only a slight conjunctivitis resulted after instillation of a 50% aqueous solution of polyallyl alcohol into the eyes of rabbits. On sacrifice of all groups at the end of 10 days, neither gross nor microscopic pathologic changes were observed. Polyallyl alcohol is a polymer of allyl alcohol, and has a molecular weight of 550 and an average degree of polymerization of 9.5. Polymerization apparently abolishes the toxic properties of the parent allyl compound.

Toxicological and pharmacological data concerning 2-benzyl-imidazoline hydrochloride (Priscol). W. BARRETT, A. CAMERON, N. HANSEN, E. HERROLD, A. MACKENZIE, B. RICHARDS, F. ROTH, J. SMITH (all by invitation), (introduced by F. F. YONKMAN). *Division of Pharmacology, Research Department, Ciba Pharmaceutical Products, Inc., Summit, New Jersey.* A continuing study of Priscol's action has yielded data which have confirmed and extended the original work of Meier and his associates (R. Meier and R. Mueller, Schwz. med. Wschr. **69:** 1271, 1939; R. Meier and R. Mueller, ibid. **71:** 1206, 1941). Toxicity: LD_{50} white rats I.V.—85–95 mg./kg.; I.P. 100–125 mg./kg., and p.o. 1200–1400 mg./kg. Dogs have tolerated orally up to 140 mg./kg. with subsequent recovery. Doses of 2.5, 5.0 and 10 mg./kg. have been administered per os 5 days a week for 60 days to dogs without revealing any significant manifestations of toxicity as determined by weight curves and hematological studies. The final results of this test, which is to continue another 30 days, will be reported. Priscol has positive, inotropic action upon isolated, perfused hearts of the guinea pig and cat. Doses from 5.0y to 2.0 mg. stimulate the former without concomitant or subsequent evidences of inhibition. Doses from 5.0 to 200y stimulate the feline heart with a slight, secondary inhibition after the highest. In the canine heart-lung preparation doses of 50 to 100 mg. exerted a mild, positive inotropic effect; lower doses exhibited no action, and doses totalling 200 mg. caused no evidences of toxicity. Warburg studies have indicated that concentrations less than 5.0 mg./cc. do not inhibit tissue respiration. The tissues studied have included renal, hepatic and cardiac slices from the rat and also feline cardiac slices. The minimal inhibitory doses varied from 5.0 to 20 y/ml. The kidney of the rat was most sensitive, and the feline heart least. The drug has revealed mild oxytocic and smooth-muscle-stimulating properties.

Effect of ion-deficient Locke's solution on guinea pig ileal responses to histamine, acetylcholine, electrical stimulation. W. BARRETT (by invitation), B. CRAVER. *Division of Pharmacology, Research Department, Ciba Pharmaceutical Products, Inc., Summit, New Jersey.* Duplicative control responses to the stimulants were elicited from the ileal strip perfused with normal Locke's solution, after which perfusion with deficient Locke's solution was initiated and the responses followed until constant. Then the effects of adding variable increments of the missing salt to the bath were determined before determining the effects of reperfusion with normal Locke's solution. The deficient solutions lacked calcium, magnesium or potassium alone or by pairs. The effects of each deficiency were reversible since sufficient reperfusion with normal Locke's solution always restored responses. The following table summarizes the results:

Missing ions	Effect on responses	Action of increments	Re-perfusion
Mg	increase	low doses—0; 1.0 mg./cc.—decrease	normal responses
Ca	absence	normal responses	normal responses
K	marked decrease	slight increase	normal responses
Ca-Mg	absence	Mg—0; Ca—normal	normal responses
Ca-K	absence	K—0; Ca—increase; K + Ca—normal	normal responses
Mg-K	absence	Mg—0; K—increase; Mg + K—normal	normal responses

The last 5 solutions caused loss of responsiveness to electrical stimulation; this was always the first stimulant to become ineffective. Intestine sensitized to horse serum and perfused with Ca-deficient solutions did not respond to the antigen until 40–80 γ/cc. of Ca had been added to bath.

Response of experimental lymphoma to certain organic sulfides, sulfones and chlorides, ALLAN D. BASS. *Pharmacology Department, Syracuse University.* It has been previously shown that bis-, (chloroethyl)sulfide is a lymphocyte destroying agent. Certain organic sulfides, sulfones and chlorides have been selected for this study in the hope of finding a less toxic agent with lympholytic action. Mouse tumor 6C3HED was used. The chemical used was administered intraperitoneally

to approximately ten C3H mice bearing the transplanted lymphosarcoma. Growth curves were plotted for each substance tested. Propylne glycol was used as a solvent or diluent where possible, but in a few instances a propylene glycol-ethyl alcohol mixture was required, and in a few instances a coconut oil emulsion was employed. Toxicities were first determined in Rockland white mice. Roughly one fourth of this dose was found to be near the minimum lethal dose for the tumor bearing C3H mice. Of the compounds studied acetodichlorohydrin proved the most effective agent. Certain problems arising from the use of solvents or diluents producing non specific tissue damage will be discussed. These factors were found to be important because of the relationship of non specific tissue damage to lymphoid tumor regression.

The effectiveness and toxicity of methadon. ROBERT C. BATTERMAN and ABRAHAM M. OSHLAG (by invitation). *Department of Therapeutics, New York University College of Medicine.* In a group of 150 hospitalized and 30 ambulatory patients, methadon in doses of 2.5 to 20.0 mg. in a single dose or in repeated doses several times daily controlled pain in only 40 per cent of the trials when administered orally and 76 per cent of the trials when used parenterally. Toxic symptoms such as dizziness, nausea, vomiting, weakness and drowsiness were frequently noted occurring in hospitalized patients in 23 and 40 per cent for parenteral and oral administration respectively and 80 per cent for ambulatory patients. When patients above the age of 50 receive the drug repeatedly for several days, severe mental confusion and toxic psychosis was noted in 30 per cent of the trials. Methadon is not a satisfactory analgesic. The high incidence of untoward reactions noted after chronic administration and in ambulatory patients limit its clinical usefulness.

Infectivity of sporozoites of Plasmodium Cathemerium 3H2 exposed in vitro to hen and canary bloods. HARRY BECKMAN. *Marquette University School of Medicine.* Mosquitoes carrying the sporozoites of *P. cathemerium 3H2* were ground in Locke's solution and added simultaneously to freshly drawn canary (susceptible) and he susceptible) bloods. The two blood-sporozoite mixtures were incubated with gentle agitation at 41.5°C., and at thirty-minute intervals through two and a half hours increments representing one mosquito each were withdrawn and injected intramuscularly into canaries, the peripheral blood of these birds being subsequently examined from the fifth to the sixteenth days, inclusive. Birds failing to become positive were challenged by mosquito bite. Trials were run at sufficient intervals that at least one complete three week sporozoite-to-sprozoite cycle lay between. Of the total of eighty canaries injected with sporozoites previously incubated in canary blood, seventy-eight (97.5%) showed patent blood infections; of the seventy-six canaries injected with sporozoites previously incubated in hen blood, two (2.6%) showed patent blood infections. All birds failing to become positive were subsequently successfully infected by mosquito bite. It would appear that the blood of the hen contains a factor capable of preventing patency of *P. cathemerium 3H2* in canaries injected with sporozoites previously incubated in it.

Studies on a heparin-like compound, algarin. ALBERT J. BEGANY (by invitation) and JOSEPH SEIFTER. *Wyeth Institute of Applied Biochemistry, Philadelphia, Pa.* Algarin is a sodium salt of an alginic acid disulfuric ester containing 15–17% sulfur. In common with other polysaccharide-polysulfuric acid esters it is a powerful anticoagulant *in vivo* as well as *in vitro*. It differs from the heretofore described substances of this class in its relatively low toxicity and failure to produce hemorrhage of the viscera. Large doses do not affect the blood pressure when injected intravenously. In this study data is presented comparing Heparin and Algarin by intravenous injection in rabbits, cats, and dogs. Algarin appears to be $\frac{1}{5}$–$\frac{1}{12}$ as active as Heparin depending on species employed. When these ratios are employed the intensity and duration of effect are practically identical for Algarin and Heparin. Algarin is not inactivated by the liver. It is antidoted by intravenous injections of protamine sulfate or toluidine blue.

Prolonged depressor effect of water soluble organic nitrates. JOSEPH G. BIRD (introduced by JOHN C. KRANTZ, JR.). *Department of Pharmacology, Univ. of Maryland, School of Medicine, Baltimore.* It has been observed that no reduction of blood pressure in the dog is produced by the intravenous injection of the sodium salt of glycollic acid nitrate in doses equal to effective amounts of glyceryl trinitrate. However, larger doses evoke a depressor response more prolonged than responses to glyceryl and erythritol nitrates. Of particular interest is the insensitivity of the lowered arterial pressure to repetition of the dose of glycollate nitrate although the foregoing nitrates still produce their transient depressor action. Malic and tartaric acids have been nitrated and are being studied. Water solubility appears to be an important factor in the action of organic nitrates.

An examination of potentiating action of aspirin upon codeine in raising the pain threshold. D. D. BONNYCASTLE and JACOB MOLLAND (by invitation). *Departments of Pharmacology, Yale School of Medicine, and University of Oslo, Oslo, Norway.* In this investigation a modification of the D'Amour-Smith radiant heat apparatus has been employed. Rats were used as the test animals,

and in carrying out this experiment special consideration has been given to certain factors which we have shown to be important in this type of work, viz.

(i) Training periods are necessary for the animals before use.

(ii) A daily cut-off time of stimulation must be established to avoid injury which might impair the animals' usefulness in the carrying out of subsequent tests.

(iii) An adequate statistical design should be used for this type of work.

Acetylsalicylic acid in dose levels that have little or no effect upon the pain threshold have been used with graded doses of codeine sulphate. The dose levels of codeine sulphate range from those that are without effect to ones having a definite action in raising the pain threshold.

Re-evaluation of the effectiveness of metrazol as an analeptic agent in barbiturate depression, WALTER M. BOOKER, DAVID M. FRENCH, PEDRO A. MOLANO, and CECIL RHODES (introduced by A. H. MALONEY). *From the Department of Pharmacology, College of Medicine, Howard University, Washington, D. C.* A growing controversy is more and more imminent as to the effectiveness of metrazol in overcoming barbiturate depression. Interesting divergent pieces of experimental evidence have been published. The increasing widespread use of pentothal sodium as an anesthetic agent has accentuated and sharpened the opposing views. We present evidence showing that metrazol is effective in stimulating respiration and even arousing animals depressed by pentothal sodium. Groups of dogs have been anesthetized by administering pentothal sodium intravenously at 20, 30, 40 and 50 mgm./kgm. body weight, and the mgm./kgm. ratio of metrazol necessary to restore normal respiration and arouse the animals at each level of barbiturate administration was determined. Figures varying from 125 mgm./kgm. body weight to 500 mgm./kgm. body weight of metrazol are presented, in relation to the varying dosage levels of pentothal administered. In a number of animals, blood pressure tracings, expired air values, and arterial oxygen saturation values have been obtained. Our evidence seems to show that although respiratory rate may be increased in most instances where high doses of metrazol are used, the depth of respiration is of much more importance in improving the condition of the animal. This is shown by oxygen saturation of arterial blood and by the state of the blood pressure.

The mechanism of the extraction of bromsulfalein from blood plasma by the liver. RALPH W. BRAUER and RITA L. PESSOTTI (introduced by CHAPMAN REYNOLDS). *Department of Pharmacology and Experimental Therapeutics, School of Medicine, Louisiana State University, New Orleans, La.* Bromsulfalein (BSP) forms compounds with various plasma proteins. With bovine plasma albumin (crystalline) (CBPA) the reaction involves one molecule of dye and one of protein, and is reversible (dissociation constant 7×10^{-6}). In human plasma most of the BSP is associated with the true albumin fraction. Liver homogenates likewise form non-dialyzable complexes with the dye. Rat liver slices incubated with low concentrations of BSP in Krebs solution take up insignificant amounts of the dye. With increasing concentrations of BSP dye uptake by the tissue occurs in such fashion that a constant BSP concentration remains in the supernatant despite considerable further increase in the initial dye concentration. Once the storage capacity of the liver slices is exceeded, all of the excess dye appears in the supernatant. The addition of CBPA to the Krebs solution reduces the fixation of BSP by the liver. In perfused rat livers the percent dye extraction from Locke's solution is almost independent of the initial concentration. Increased perfusion rates, or the addition of CBPA to the perfusion fluid reduces the proportion of BSP extracted. Continuous i.v. BSP infusion (normal dogs) produces steady plasma and bile levels, biliary excretion at 70–100% infusion rate. In each animal a constant proportion (60–85%) of the dye is removed from blood passing through the liver, regardless of the systemic plasma level. These and related results obtained under various abnormal conditions indicate a minimal participation of the RE system in the extraction of BSP from plasma. Instead, an exchange of protein bound dye between plasma and hepatic cells is proposed as the basic mechanism.

The physiological disposition of acetophenetidin (p-ethoxyacetanilide) in man. BERNARD B. BRODIE and JULIUS AXELROD (by invitation). *Department of Biochemistry, New York University College of Medicine, and New York University Research Service, Goldwater Memorial Hospital, and Laboratory of Industrial Hygiene, N. Y., N. Y.* Previous work concerning the metabolism of acetanilide (unpublished) has shown that acetanilide deacetylates in small part in the body to aniline, the precursor of the substance which oxidizes hemoglobin to methemoglobin, and, in large part, to N-acetyl p-aminophenol, an active analgesic. The metabolism of acetophenetidin has also been studied as part of a program concerning the fate of aniline derivatives in the body. The route of metabolism of acetophenetidin was shown to be as follows: A minor fraction of the drug deacetylates to form p-phenetidin (p-ethoxyaniline). This compound was shown to be the precursor of the substance which oxidizes hemoglobin to methemoglobin. The major fraction of the drug deethylates rapidly to form N-acetyl p-aminophenol.

The short life of the parent drug in the body and and its rapid alteration to N-acetyl p-aminophenol leads to the conclusion that the analgesic activity of acetophenetidin is due to this derived product. Preliminary work with N-acetyl p-aminophenol indicates that it is an effective analgesic and may be given frequently in relatively large doses without producing methemoglobin.

Quantitative epinephrine pressor effects. ROBERT V. BROWN. *Division of Pharmacology, University of Tennessee, Memphis.* Past experiments on the pressor responses to epinephrine have produced dose-effect curves defined as linear, logarithmic, hyperbolic, or exponential. The effects were measured as rises in blood pressure by some, as peak blood pressures by others. Hjort, et. al., (J. Pharmacol. & Exper. Therap. **71:** 105, 1941) fit their data best with the equation $\log y = b \log x + \log a$, where y is the increase in blood pressure and x is the dose of epinephrine. Their data were averaged and a single curve was published. In a series of experiments in dogs, pressor effects were measured both as blood pressure increases and as peak blood pressures. Both sets of effects follow the log dose-log effect regression line, but the log dose-log peak equation, determined by the least squares method, yields a much better fit to the experimental data than the log dose-log increase equation. When the dose is in μgm./kgm., the constant, log a, is the intercept where $x = 1$ or $\log x = 0$. The constant, log b, is the slope of the regression line. Both constants are measures of the sensitivity of any particular animal to the pressor effects of epinephrine. They vary from animal to animal and each has two values, depending on whether increases in blood pressure or peak blood pressures are used. It is possible to fit this equation to various already published data. The previously proposed equations diverge continuously from the experimental data as the lower range of dosage is traversed. The most accurate dose-effect curve, where Y is the peak blood pressure and x is in μgm./kgm. is $Y = Ax^B$ or $\log Y = B \log x + \log A$.

On the mechanism of the antitussive action of amidone. MARION BUELL (by invitation), JOAN E. COPELAND (by invitation) and ELDON M. BOYD. *Dept. of Pharmacology, Queen's Univ., Kingston, Canada.* The morphine-like action of amidone has naturally suggested its trial in the symptomatic therapy of cough due to the common cold and a number of other conditions. If it be accepted that amidone is an effective antitussive drug, the question arises as to whether it acts by increasing the secretion of bronchial or respiratory tract fluid (R.T.F.), or by centrally depressing the cough reflex, or in some other manner. The first mode of action noted has been investigated using a modification of the technique of Perry and Boyd (J. Pharmacol. & Exp. Therap., **73:** 65, 1941). Experiments were performed upon several species of animals, the drug being given both orally and parenterally in a series of doses ranging from 0.1 to 10 mgm. per kilo body weight. In no species could there be demonstrated a statistically significant change in the volume output of R.T.F. In this negative pharmacodynamic action, amidone apparently acts like morphine which also has been shown to have no effect upon the volume output of R.T.F. (Boyd and MacLachlin, Canad. Med. Assoc. J., **50:** 338, 1944).

Pharmacological studies of myanesin and curare. JOHN C. BURKE (by invitation) and CHARLES R. LINEGAR. *Pharmacological Development Division, E. R. Squibb & Sons, New Brunswick, New Jersey.* The pharmacological action of myanesin, 3-(o-toloxy)-1,2-propanediol, has been compared with that of d-tubocurarine chloride in various animals. Myanesin produces muscular relaxation similar in appearance to that of curare. Degree of relaxation with both drugs is dependent upon dosage, and complete relaxation of postural muscles can be accomplished without paralysis of respiratory muscles. This paralysis can be used as the "head-drop" end point in the assay or the comparison of such drugs. Onset of paralysis with myanesin is more gradual in response to dose than with curare. Partial paralysis by myanesin is characterized by incoordination, stupor and partial analgesia, while curare produces only simulated muscle weakness. Myanesin occasionally causes transient rigidity, frequently nystagmus at the peak of paralysis, and nausea and vomiting during recovery. These reactions are absent with curare. Myanesin shows a wider species difference in potency than curare but approximately the same therapeutic ratio. d-Tubocurarine chloride is 230 to 1,000 times as potent as myanesin intravenously in different species while the duration of action of the two drugs is similar. Myanesin exhibits some local anesthetic activity; curare none. Myanesin causes severe vasodepression at relaxing dosage, and severe cardiac depression or block at respiratory paralyzing dosage. It also causes significant hemolysis *in vivo* and *in vitro*. The paralyzing action of myanesin is due to depression of the central nervous system, reaching above the spinal level, and to some slight peripheral or myoneural depression, while that of curare is peripheral, being accomplished without significant vasomotor, cardiac or central nervous system depression.

A comparison of the effects of ether and cyclopropane on renal function in man. BURNETT, CHARLES H. (by invitation), GORDON, ESTHER B. (by invitation), SHORTZ, GERALD (by invitation), COMPTON, DAVID W. (by invitation) and BEECHER, HENRY K. *Anesthesia Laboratory of the Harvard Medical School at the Massachusetts General Hospital and the Department of Medicine, Massachusetts General Hospital.* The effects of anesthesia

upon renal clearance of mannitol and sodium para-amino-hippurate in 8 patients under ether and 7 under cyclopropane have been compared. Only patients in good clinical condition with essentially normal renal function were chosen, and all studies were completed before surgical procedures were started. Anesthesia was maintained in the second plane of the third stage. Average observed changes are shown in the table. Individual protocols reveal

Anesthesia	No. patients	Average changes (per cent)			
		C_M	C_{PAH}	C_M/C_{PAH}	Urine flow
Ether	8	−21.2	−38.7	+25	−45
Cyclopropane	7	−32.5	−53.3	+35	−42

some trends and differences not apparent from mean values. One subject showed no significant change in either mannitol or para-aminohippurate clearance during ether; in a few other ether subjects clearance of both substances following an initial drop soon after induction improved as anesthesia continued. This trend toward improvement was absent or much less marked in the cyclopropane group. Carbon dioxide contents of the serum fell slightly with ether and were unchanged with cyclopropane. In so far as could be determined, extrarenal factors were excluded or so minimized that the observed changes in glomerular filtration rate and effective renal plasma flow represent effects on the kidneys *per se* of these agents. The high filtration fractions suggest that the renal ischemia is at least partly due to efferent arteriolar constriction. Our data indicate that while qualitatively the effects of ether and cyclopropane on renal function are similar, quantitatively those of cyclopropane are more marked in reducing renal function.

Actions and minimally effective doses of 17 common drugs on perfused hearts of 5 species. A. CAMERON (by invitation), M. LASKEY (by invitation), B. CRAVER. *Division of Pharmacology, Research Department, Ciba Pharmaceutical Products, Inc., Summit, New Jersey.* Since a standard method for studying the responses to drugs of isolated, perfused hearts had become available (F. F. Anderson and B. N. Craver, Fed. Proc. **6**: 48, 1947), it seemed valuable to prepare a table recording the quantitative and qualitative differences in responses to common drugs of the hearts of the usual laboratory animals. Such a table as the following would not only be a convenience to those employing the Langendorff preparation, but might provide clues to modes of drug action and cardiac metabolism.

Drug	Cat	Rabbit	Guinea pig	Rat	Hamster
Acetylcholine	1.0y −	0.1y +	0.005–0.01y ±	0.005y −	0.005y −
Aminophylline	100–200y + −	100–200y −	10–50y +	5.0y −	3.5y −
Antistine	20–50y −	20–50y −	5.0y −	5–10y −	10y −
Atropine*	20y −	20–50y −	10–20y −	1–2y −	0.5y −
Digifoline	0.025–0.05 c.u. +	0.05 c.u. +	0.025 c.u. +	0.05–0.1 c.u. +	0.1 c.u. +
Doryl	0.5y −	0.25–0.5y +	0.001–0.005y ±	0.002y −	0.002y −
Ephedrine	0.5–1.0y +	4.0y −	0.5y +	2.0y–5.0y +	1.0–2.0y −
Epinephrine	0.019y +	0.01–0.02y +	0.01–0.02y +	0.01–0.02y +	0.02y +
Histamine	variable	0.2y −	0.005–0.01 y ±	5.0y −	10–20y ±
Mecholyl	1.0y −	0.5–1.0y +	0.002–0.005y +	0.005y −	0.005y −
Papaverine	50–100y +	50–100y −	2.0–5.0y +	5–10y +	0.5y −
Pituitrin (S)	variable	0.002 I.U. +	0.002–0.004 I.U. +	0.001–0.002 I.U. +	0.01–0.02 I.U. −
Priscol	5–20y +	200–300y −	5.0y +	10y −	10–20y −
Privine	20–50y −	10y −	0.5y −	1.0y −	0.5–1.0y −
Pyribenzamine	20–50y −	50y −	5.0y −	5–10y −	10y −
Tetraethyl ammonium bromide	variable	variable	20y −	10y −	3.0–5.0y −
Trasentine	20–100y −	50–100y −	5–10y −	3.0y −	5–10y −

* This dose mildly inhibits cardiac action, but lower doses (2–3y) block effective doses of Doryl and Mecholyl. + = stimulation. − = inhibition.

The use of ultraviolet rays in a screening test for some barbiturates and in reading Gutzeit papers. W. J. R. CAMP, V. A. GANT (by invitation) and JOHN F. POLLI (by invitation). *Dept. of Toxicology and Pharmacology, Univ. of Ill. College of Medicine, Chicago 12.* When a large number of

toxicological analyses are to be made, any procedure which will expedite or simplify the procedures is of importance. The number of substances dispensed in colored capsules is rather limited, and from the viewpoint of toxicology really limited to a few very commonly used barbiturates. Occasionally one may find remnants of colored capsules in the stomach contents. The dyes used in the capsule are not sufficient to distinctly color the specimen but are in sufficient quantity to reveal a very definite fluorescence when exposed to filtered ultraviolet rays. While this procedure is not indicative of barbiturates, since other materials, e.g., Benadryl, are dispensed in colored capsules, it is helpful in shortening chemical procedures when a history is wanting and the fluorescence is positive. Naturally the absence of a fluorescence should not exclude analysis for barbiturates. A second use for the ultraviolet lamp is for a more accurate measuring of the stain on Gutzeit paper. A golden glow is seen to sharply limit the area of unchanged mercuric bromide. The strip is marked under the light and compared with standard measurements similarly made. We have found that strips negative to the eye show a definite fluorescence. While there is a slight increase in the sensitivity we feel the ease in accurately reading the strip is of more importance.

The acute toxic effects of dicumarol. Rose Marie Carlson (by invitation) and Lloyd D. Seager. *Department of Pharmacology, The Womans Medical College of Pennsylvania.* The observations of Rose, Harris and Chen, Proc. Soc. Exp. Biol. and Med., **50:** 228, 1942 on the toxic effects of dicumarol have been largely confirmed. In addition a direct toxic effect of the drug on the heart has been found. In the cat, rabbit, rat and mouse the intravenous or intraperitoneal injection of 100 to 300 mgs. per kg. produces death within a few hours. Death is preceded by convulsions. Animals biopsied during convulsions have invariably shown cardiac arrest usually involving both auricles and ventricles and often presenting fibrillation. Blood pressure tracings show cardiac acceleration with little change in pressure until the terminal abrupt fall. The frog also presents cardiac arrest after the lymph sac injection of 200 mgm. per kg. It is suggested that the convulsions from large doses of dicumarol are asphyxial in nature and that the pulmonary edema frequently encountered is in part cardiac in origin.

The LD_{50} of pentobarbital in nursed and unnursed newborn rats. Emmett B. Carmichael and Walter H. Johnson (by invitation). *Biochemistry Department, Medical College of Alabama, Birmingham.* Young rats less than 24 hours old were used. All injections were made intraperitoneally and the animals were immediately placed in an incubator to avoid chilling. The stoppage of the heart beat was taken as the time of death. The doses employed were 30, 40, and 50 mg./kg. body weight. The rats were separated into three weight-groups: (1) 4.0–4.99 grams, (2) 5.0–5.99 grams, and (3) 6.0–7.5 grams. The nursed rats survived a larger dose than did those that did not nurse. The LD_{50} for the nursed rats seems to be almost 40 mg./kg. while the LD_{50} for those that did not nurse was about 30 mg./kg.

A comparative study of cyclic and noncyclic hydrocarbons on cardiac automaticity. C. Jelleff Carr and John C. Krantz, Jr. *Department of Pharmacology, Univ. of Maryland, School of Medicine, Baltimore.* Muth, Hathaway and Orth (1937) showed that arrhythmias frequently occurred in the dog's heart under cyclopropane anesthesia when the automatic tissue of the heart was sensitized by intravenous injections of epinephrine. In this laboratory it has been shown that the *Macacus rhesus* monkey behaves in a similar manner and that cyclobutane anesthesia sensitizes the dog's heart to epinephrine similar to cyclopropane anesthesia. In the present study the effects of other hydrocarbons related to cyclobutane and cyclopropane have been investigated. Anesthesia in the dog under cyclobutane, cyclobutene, butane, isobutane and *cis-trans* 2-butene sensitizes the automatic tissue of the myocardium to epinephrine. Anesthesia under ethylene does not elicit this response. Cyclobutane and cyclobutene produce an excellent anesthesia in the dog; butane, isobutane and *cis-trans* 2-butene do not evoke satisfactory anesthesia in the dog.

The anaphylactic guinea pig trachea and its response to antihistamine and bronchodilator drugs. Julio C. Castillo (introduced by Edwin J. de Beer). *The Wellcome Research Laboratories, Tuckahoe, 7, New York.* In a paper published in the Journal of Pharmacology and Experimental Therapeutics, **90:** 104, 1947, a new method is described for magnifying and recording the constrictions and dilatations of the trachea of the guinea pig when exposed to drugs. The technique consists in sectioning the entire trachea into 12 rings of approximately equal thickness and connecting the rings in chain-fashion with short loops of thread. The "tracheal chain", as the preparation is called by the authors, is mounted in a tissue bath and connected to a light muscle lever for kymographic recording. "Tracheal chains" made from guinea pigs sensitized to horse serum were found to respond to small concentrations of the antigen with marked contractions which were maintained at the same level for a long period of time. In view of the close anatomical and physiological relation which exists between tracheal and bronchial musculature, these anaphylactic constrictions of the trachea promise to be very useful in the study of drugs which may find application in the relief of certain types of allergic asthma. The anaphylactic spasm, as produced in the guinea pig trachea, dif-

fers from that obtained with histamine mainly in the fact that it cannot be relaxed by either washing nor by the use of antihistamine drugs. Bronchodilator drugs, on the other hand, were found to be very effective in relieving the spasm. These results stir up interesting speculations as to the role that histamine may play in allergic asthma.

Prolongation of curare action with a peanut oil and beeswax vehicle. HAROLD F. CHASE and BANI K. BHATTACHARYA (by invitation). *From the Departments of Pharmacology and Surgery, Western Reserve University and The University Hospitals of Cleveland, Cleveland, Ohio.* The efficacy of a beeswax and peanut oil vehicle (Romansky formula) in delaying the absorption of d-tubocurarine has been studied by the rabbit head-drop method of assay. Control determination of head-drop doses was made in a group of nine rabbits using a preparation containing 0.5 mgm. of crystalline d-tubocurarine per cc. of distilled water. This preparation was used to titrate the same animals 24 and 48 hours subsequent to the subcutaneous injection of 4.0 mgm. per kgm. of d-tubocurarine in beeswax and peanut oil, containing 30 mgm. per cc. The 34.4 per cent and 10.5 per cent reductions in head-drop doses, found, respectively, 24 and 48 hours after the oil injections, were statistically significant and were assumed to indicate the activity of the residual effective curare in the animal. This evidence would suggest that prolonged muscular relaxation of spastic patients following injection of this preparation may be due to true curare action rather than to what has been termed "lissive" action.

The joint antihistaminic effect and acute toxicity of adrenalin and benadryl·HCl. GRAHAM CHEN, A. C. BRATTON, JR. and CHARLES ENSOR (by invitation). *Research Laboratories, Parke, Davis* M *Company, Detroit, Michigan.* In histamine-induced bronchospasm of guinea pigs, the 50 per cent protective doses of Adrenalin (A), Benadryl·HCl (B), and the combination of the two (A + B) were determined by aerosol treatment as follows: A = 45.31 ± 5.91 γ/L, B = 11.56 ± 1.41 γ/L and A + B = $[21.87 \pm 2.69\ \gamma\ (A) + 5.63 \pm 0.78\ \gamma\ (B)]$/L. The LD_{50}'s of the compounds by intramuscular injection in mice were found to be 2.93 ± 0.15 mgm./kg. for (A), 58.10 ± 2.21 mgm./kg. for (B) and 1.20 ± 0.08 mgm./kg. (A) plus 22.00 ± 1.52 mgm./kg. (B) for the combination. Toxic doses of Benadryl·HCl produced an increase in blood glucose from 30 to 100 per cent in rabbits. The hyperglycemia was caused by central nervous stimulation and could be prevented by the administration of pentobarbital. Benadryl·HCl and Adrenalin given together exerted independently their hyperglycemic effect. The results indicate that the joint antihistaminic and hyperglycemic effect of Adrenalin and Benadryl·HCl is additive while the joint lethal toxicity of the two at low dosages of Benadryl·HCl is synergistic in nature.

The electrogram of the papillary muscle. MAYNARD B. CHENOWETH and SOLOMON GARB (by invitation). *Dept. of Pharmacology, Cornell Univ. Medical College, New York, N. Y.* An isolated cat papillary muscle was prepared following the method of Cattell and Gold. An additional pair of electrodes recorded the electrogram of the muscle. Studies were made of the effects of fatigue, injury, and rate, and polarity changes. In addition, the effects of such drugs as epinephrine, acetylcholine, chloroform, digitoxin, veratrine, quinidine, caffeine, and procaine on the electrogram and myogram were studied.

Effect of some derivatives of subtilin on tubercle bacilli and rabbit leukocytes in vitro. YIN-CH'ANG CHIN (introduced by HAMILTON H. ANDERSON). *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco, California.* A number of derivatives of subtilin have been prepared by the Western Regional Research Laboratory at Albany, California. The bacteriostatic action of the methylated, ethylated, and glycolated derivatives on *Micrococcus conglomeratus*, *Staphylococcus aureus*, and *Streptococcus faecalis* have been found to be three times that of the original lots of subtilin (personal communication). The original lots and derivatives were submitted to this laboratory for the study of their bacteriostatic action on tubercle bacilli and toxicity to rabbit leukocytes. The bacteriostatic concentrations were determined on human tubercle bacilli, strain H37Rv, in Dubos' fluid medium. This strain was more resistant to subtilin than the strain R1 (reported by the author in Federation Proceedings, 6: 317, 1947) which was isolated from an active case of pulmonary tuberculosis by Dr. G. E. Rockwell in 1937 (personal communication) and supplied to this laboratory through the Lilly Research Laboratories at Indianapolis, Indiana. Rabbit leukocytes were obtained by washing them from the peritoneal cavity and the appropriate dilutions of subtilin and its derivatives were made from their solutions in physiologic saline. The results are shown in the accompanying table. It would appear that methylated derivatives were twice as active as the original samples against tubercle bacilli while

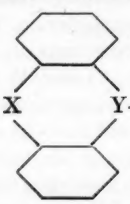
Lot no.	Modification of subtilin		Bacteriostatic concentration against H37Rv (in Dubos medium)	Concentration in which leukocytes showed no apparent change after 60 minutes at 37°C.
114-Q	—	—	1:200,000	1:1,000
L1318	—	—	1:100,000	1:1,000
L1319	CH_3	L1318	1:200,000	1:1,000
L1322	—	—	1:100,000	1:1,000
L1329	CH_3	L1322	1:200,000	1:3,000
L1330	CH_3	L1322	1:200,000	1:1,000
L1331	C_2H_5	L1322	1:100,000	1:1,000
L1332	C_2H_4	L1322	1:100,000	1:1,000

only one of them showed an increased toxicity to leukocytes.

Effects of surface active agents on the activity of pentobarbital and barbital. V. V. COLE, S. H. HOPPER (by invitation), and H. R. HULPIEU. *Depts. of Pharmacology and Public Health, Indiana Univ. School of Medicine, Indianapolis, Ind.* Mixtures of eleven different surface active agents with a hypnotic dose of sodium pentobarbital were injected intravenously into mice and compared with mice receiving only sodium pentobarbital. In each case the sleeping time was longer with a surface active agent than with sodium pentobarbital alone. The increase in sleeping time varied from 42 to 105 per cent. Seven of these surface active agents were given 5 minutes before sodium pentobarbital and a significant difference from the mixtures was found with only one agent, which gave a greater increase when given before sodium pentobarbital. Another agent (Aresklene 400), which gave the greatest increase by the first method and essentially the same results by the second method, was chosen for study with other animals and with sodium barbital. This agent caused an 82 per cent increase in average sleeping time in rats. Rabbits showed no change in sleeping time but non-fatal doses of sodium pentobarbital proved fatal given with Aresklene 400. In dogs there was a decrease in sleeping time. With sodium barbital as the hypnotic agent there was no difference in sleeping time in the species tested. The time elapsing from injection of sodium barbital to onset of hypnosis was markedly less with Aresklene 400 in mice, rabbits, and dogs while there was no difference in rats.

Effect of anti-cholinesterases upon procaine toxicity. A. C. CONWAY (by invitation), F. S. TING (by invitation), and JULIUS M. COON. *Department of Pharmacology, University of Chicago.* An enzyme present in blood serum capable of destroying procaine was described in 1943 by Goldberg et al. Kisch demonstrated in vitro that this enzyme was not a cholinesterase but that, like cholinesterase, it could be inhibited by physostigmine and neostigmine. In view of this it was thought of interest to determine what influence these anti-cholinesterase agents might have on the toxicity of procaine. Studies were conducted on mice and rats. The mice were first injected subcutaneously with physostigmine salicylate (0.3 mg./kg.) or neostigmine methyl sulfate (0.2 mg./kg.). Procaine hydrochloride was administered intravenously via the tail vein about ten minutes later. In the case of physostigmine the LD50 for procaine rose from 56 mg./kg. to 66 mg./kg., thereby indicating a slight protective action of physostigmine upon procaine toxicity. The severity of convulsions due to procaine were also markedly diminished. This same effect was observed in rats and dogs. When neostigmine was tested with procaine an opposite effect on toxicity was noted. The LD50 for procaine was reduced to approximately 30 mg./kg. Deaths in these instances were practically instantaneous and no convulsant phase occurred. The failure of physostigmine to increase the toxicity of procaine indicates that some factor other than its antiprocainesterase effect operates to influence toxicity. It appears that this mechanism is lacking in the case of neostigmine. This pharmacological difference between physostigmine and neostigmine is being tested in other animal species.

Inhibition of histamine hypotension by certain heterocyclic substituted ethylamines in the cat. By DONALD L. COOK (by invitation), W. E. HAMBOURGER, and MARTIN M. WINBURY (by invitation). *From the Pharmacology Laboratories, Research Department, G. D. Searle & Co., Chicago, Ill.* The method described by Wells *et al.* (J. Pharmacol. **85:** 122, 1945) for calculating inhibition of the histamine depressor response in dogs has been used to compare the activities of six compounds with diphenhydramine HCl in cats. These compounds



have the general formula, $N(CH_3)_2 \cdot Z$, where X is S, SO, or SO_2; Y is N or CH; and Z is HCl or 8-chlorotheophylline. Preliminary experiments indicated that for each compound histamine antagonism persisted with only slight diminution for at least one hour, permitting the assumption that repeated doses would be cumulative. Three to six animals were, therefore, given doses ranging from 0.025 to 3.2 mg./kg. on a geometric scale. In each experiment the hypotension produced by logarithmic doses of histamine diphosphate from 0.125 to 16.0 gamma/kg. was determined before and after each successive dose. From these data, per cent inhibition at each dose level was determined. Curves are obtained when per cent inhibition is plotted against dosage. This curve for DPH in cats is very similar to Well's curve for DPH in the dog. The data may also be plotted to yield a family of parallel straight lines from which the relative activities of the compounds can be derived. The HCl and 8-chlorotheophylline salts of dimethylaminoethylphenothiazine are the most active compounds in the series.

A pharmacologic comparison of hexaethyl tetraphosphate (HETP) and tetraethyl pyrophosphate (TEPP) with physostigmine and neostigmine. JULIUS M. COON and PAUL R. SALERNO (by invitation). *University of Chicago Toxicity Laboratory and the Department of Pharmacology, University of*

Chicago. Hexaethyl tetraphosphate and tetraethyl pyrophosphate have been reported to be potent cholinesterase inhibitors. A study of the effects of HETP and TEPP on the circulatory system of the cat and dog and on the isolated heart, intestine and uterus of the rabbit showed that these agents are largely similar in action to physostigmine and neostigmine. In anesthetized cats and dogs HETP and TEPP elicited a marked pressor action followed by a cardiac slowing which was prevented or abolished by atropine or procaine but not by vagotomy. The pressor effect was not influenced by nicotinization or adrenalectomy but could be abolished by large doses of dibenamine-HCl. The isolated heart was relatively insensitive to these drugs, tolerating without effect a 1:10,000 concentration in the perfusing fluid. Larger quantities depressed the activity of the heart. This depression was not affected by atropine. Inserted into the perfusion line, the first dose caused a decrease in coronary flow and subsequent doses caused an increase. The isolated intestine was highly sensitive to these agents. Concentrations of 1:10,000,000 HETP and 1:100,000,000 TEPP, after a latent period, abolished the pendulum movements and established a peristaltic rhythm. This change could be reversed by atropine, procaine, thiamine and nicotine, but not by repeated washing. The similar effects of physostigmine and neostigmine were easily reversed by washing. The cat and rabbit intestine in situ were also very sensitive to the intravenous injection of HETP and TEPP. The isolated uterus was insensitive to these drugs.

Total body water by D_2O tracer study in protein depletion. F. Co Tui, V. Hollander, A. S. Keston, J. H. Mulholland. *Department of Surgery, New York University College of Medicine.* The results of a study of the total body water of depleted patients determined by the use of D_2O as tracer will be presented.

Protein depletion syndrome and responses to hyperalimentation. F. Co Tui, J. H. Mulholland, N. Kuo, *Department of Surgery, New York University College of Medicine.* The plasma volume and related perimeters were determined in a series of patients with marked weight loss resulting from disease. Significant deviations from normal were found. Some of these patients were then placed on hyperalimentative intake and the evolution of these perimeters toward normal was followed.

Pharmacological actions of 2-phenylbenzylaminomethyl-imidazoline (antistine). B. Craver, W. Barrett (by invitation), A. Cameron (by invitation), H. Hays (by invitation), G. Holmquist (by invitation), A. Mackenzie (by invitation), J. Smith (by invitation). *Division of Pharmacology, Research Department, Ciba Pharmaceutical Products, Inc., Summit, New Jersey.* The following additional data have been secured for Antistine (as the hydrochloride, sulfate or methane sulfate) which was initially prepared by Miescher, Urech and Klarer, and originally studied pharmacologically by Meier and Bucher (Schweiz. med. Wschr. **76:** 294, 1946.). The intravenous LD_{50} for white rats is 39 mg./kg. and, as with other antihistaminics, convulsions precede death. Rats tolerated 100 mg./kg. per os 5 days a week for 30 days with no evidences of toxicity as judged by weight curves and hematological studies. Rats given 10 mg./kg. per os 5 days a week for 30 days did not evidence tolerance or accumulative toxicity as judged by subsequent responses to the previously determined LD_{33} and LD_{83}. Doses of 10 to 50y inhibited isolated, perfused hearts. That of the guinea pig was most sensitive; the hearts of the rabbit and cat least sensitive. The highest concentration that did not inhibit ciliary activity was 1:1000. Five preparations have evidenced the potent antihistaminic properties of this drug: (1) Doses of 100 y/kg. markedly inhibited, or briefly eliminated the salivary flow induced by the intra-carotid injection of histamine in cats. Lower doses inhibited histamine's sialogogic effect; (2) doses from 1.0 to 5.0 mg./kg. diminished histamine's hypotensive action from 30 to 40% in dogs; (3) Doses from 1.5 to 3.0 mg./kg. eliminated the intestinal spasm induced by intravenously injected histamine (10-20 y/kg.) in "Thiry-Vella" dogs; (4) 0.1 y/ml. eliminated the stimulating action of 1.0 y/ml. of histamine diphosphate upon isolated ileal strips from the guinea pig; (5) Lungs of the guinea pig perfused with solutions containing 1.0 to 10 y/ml. became unresponsive to histamine. These data commend the clinical use of the drug. Its therapeutic ratio is high and akin to that of PBZ since Antistine is from $\frac{1}{3}$ to $\frac{1}{10}$ as active antihistaminically, and about $\frac{1}{3}$ to $\frac{1}{4}$ as toxic.

A comparison of the fat storage of the isomers of benzene hexachloride in rats. Bernard Davidow, Ernest C. Hagan and Geoffrey Woodard (introduced by Arnold J. Lehman). *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington 25, D. C.* Although in single oral doses the toxicity of the isomers of benzene hexachloride decreases in the order gamma, alpha, delta, and beta, it has been observed in this laboratory that in chronic toxicity experiments the beta isomer is the most toxic. An explanation of this phenomenon was sought by investigating the relative degree of accumulation of each of the isomers in the fat of mature male and female rats after various intervals of feeding on diets containing 100 and 500 ppm. of the isomers. The fats were analyzed by a method based upon the conversion of the isomer to 1,2,4 trichlorobenzene and its estimation with an ultraviolet spectrophotometer. Striking differences in the fat storage of the isomers were found on both the 100 ppm. and the 500 ppm. diets. By far the greatest accumulation was found with the beta

isomer. Storage levels of each of the isomers were observed to be higher in females than in males. Female rats fed the 100 ppm. diets for two weeks exhibited 0.5 and 0.3 mg. of the beta isomer per gram of fat, 0.11 and 0.04 of the alpha isomer and no measurable quantities of the gamma and delta isomers. Similarly, after 12 weeks on 500 ppm. diets, female rats showed the following concentrations in the fat in milligrams per gram fat: beta, 7.5; alpha, 2.4; delta, 0.5; and gamma, 0.4.

Failure of anti-histaminic drugs to antagonise the anemia produced by fat plus choline. JOHN EMERSON DAVIS. *University of Arkansas School of Medicine, Little Rock, Ark.* Anemia was produced in four dogs by the daily feeding of choline chloride and fat (lard) according to the technique described previously (Am. J. Physiol., **142:** 213, 1944; Science, **105:** 43, 1947). The basic diet consisted of Purina Dog Chow, which we have found effective in maintaining dogs in good health for a number of years. Clarkson and Best (Science, **105:** 622, 1947) reported failure to produce anemia with either choline alone or choline plus fat. They used an entirely different diet and therefore did not repeat our experiment. Since benadryl and pyribenzamine have been shown to have some anticholinergic potency, and since atropine has been shown to cause remission of our choline-induced anemias, it seemed desirable to determine whether the anti-histaminic drugs would antagonise the anemia caused by fat and choline. Therefore, commencing on the third experimental day when the erythrocyte counts had been reduced by one million or more, either benadryl or pyribenzamine was administered orally to the dogs in daily doses of 50 mgm. The anemias persisted in spite of the administration of the anti-histaminic compounds, showing that the latter are not as effective as atropine in this particular anemia-blocking action.

Some observations on the non-specific choline esterase in the human female. M. EDWARD DAVIS (by invitation), NICHOLAS W. FUGO, and EVELYN AI-FENG YU (by invitation). *From the Department of Obstetrics and Gynecology and the Department of Pharmacology, The University of Chicago and The Chicago Lying-in Hospital.* The non-specific choline esterase of sera was studied in pregnant, non-pregnant and gynecologically abnormal women by means of the direct titration method. The following observations were made. There is wide variation of the values obtained from individual to individual in both the pregnant and non-pregnant group. Each individual, however, showed constant values over long periods of time. There was no observable increase in non-specific choline esterase titer during gestation. Values for the non-pregnant group appear slightly higher than those of the pregnant group although both groups showed the same range of variation. Abnormally low values were obtained in the 2 cases of abruptio placentae studied. These values returned to normal postpartum and remained high for a period of 3 months. Anemia resulting from hemorrhage or from other causes had no effect on the enzyme titer.

Speed of action of thevetin after intravenous injection in man. CONRADO DAYRIT (by invitation), EDUARDO FARACO (by invitation), NATHANIEL T. KWIT (by invitation), WALTER MODELL, and HARRY GOLD. *Department of Pharmacology of Cornell University Medical College, Cardiovascular Research Unit of the Beth Israel Hospital, and Cardiac Service of the Hospital for Joint Diseases, New York.* Interest was revived by Chen and collaborators in the possible applications of thevetin in cardiac therapy. Since no clear evidence exists concerning its proper place in the use of the digitalis series, we re-examined the problem. Patients with heart failure and auricular fibrillation were placed at rest in bed. After the apex rate and the patient's condition settled down to a constant level, the effect of thevetin was tested either orally or intravenously, frequently by both methods in the same patient. The change in the apex rate was the chief guide to the effect of the drug, but it is well known that the decline of the rate follows closely clinical improvement in the congestive failure. The results show that thevetin is not satisfactory for oral administration. Large single doses of 12 to 30 cat units showed irregular and sometimes negligible therapeutic effects. This may be due to either poor absorption and/or rapid elimination. The most extraordinary characteristic of thevetin is the speed with which the effects are developed after intravenous injection; the full effect of a dose of from 3 to 6 cat units is developed in about 5 minutes (in typical cases, decline of apex rate from 140 to 100, 120 to 88, 140 to 45). This property may prove of considerable interest in the digitalization of acute heart failure in which a delay of an hour or so may prove decisive. The rapid elimination (about 6 hours) adds a factor of safety.

A comparison of side actions and analgesic effects of morphine, amidone, (4-4-diphenyl-6-dimethylamino-heptanone-3) and its isomers in man. JANE E. DENTON (by invitation), OLIVER H. STRAUS (by invitation), WILLIAM E. WADDELL (by invitation) and HENRY K. BEECHER. *From the Anesthesia Laboratory of the Harvard Medical School at the Massachusetts General Hospital, Boston, Massachusetts.* The symptoms and signs, the analgesic effects, and the effects on pulse, respiration and blood pressure produced by a single subcutaneous injection of morphine sulfate (15 mg./150 lb.), dl-amidone (10 mg./150 lb.), l-amidone (5 mg./150 lb.), iso-amidone (10 mg./150 lb.) and normal saline (1 cc./150 lb.) have been compared in a group of 29 healthy, young ambulatory men, each of whom received each of the five injections at

weekly intervals and was observed for at least five hours following each injection. The doses were chosen for equivalent analgesic effect and for amount necessary to produce sufficient incidence of side actions for statistical analysis. The identity of all drugs was unknown to the subjects and to all except one of the investigators. On the basis of symptom frequencies, times of onset and durations of symptoms, the correlation between morphine and dl-amidone, morphine and l-amidone, and dl-amidone and l-amidone varied between $+0.609 \pm 0.144$ and $+0.904 \pm 0.033$, indicating that these three drugs have virtually the same toxicity. Iso-amidone, in contrast, had zero correlation with morphine and a correlation with saline of $+0.948 \pm 0.028$, indicating that its toxicity is extremely low. Morphine, dl-amidone, and l-amidone produced a statistically significant and equal depression of respiratory rate. Iso-amidone produced slight respiratory depression, but much less than the other 3 drugs. Pulse rate was slowed to about the same degree by all drugs. No significant changes in blood pressure were observed. This Study clearly shows that morphine sulfate, dl-amidone, and l-amidone have indistinguishable side effects at dose ratios of 15 to 10 to 5 mg./150 lb. of body weight respectively, and that all side effects of dl-amidone are carried by the laevo component. The Wolff-Hardy method for pain threshold used in this study did not prove satisfactory in untrained subjects. However, our clinical study of morphine, dl- and l-amidone indicates equal analgesic effect of these doses. The analgesic effect of iso-amidone is now being assayed clinically.

Procaine as an antiallergic agent. ROBERT H. DREISBACH and NAI CHU (Fellow, American Bureau for Medical Aid to China Inc.; by invitation). *Dept. of Pharmacology and Therapeutics, Stanford University School of Medicine, San Francisco 15, Calif.* Recent reports stressing the use of procaine intravenously in the treatment of serum sickness and urticaria prompted a trial of procaine against experimental sensitizations and histamine toxicity. Guinea pigs were sensitized by the subcutaneous injection of 0.1 cc. of horse serum. Three weeks later, 12 of these were injected with 50 mg./kg. of procaine subcutaneously. Thirty minutes later, 0.5 cc. of horse serum was injected intravenously. Eleven animals died of typical anaphylactic shock. At the same time, 11 sensitized animals were injected intravenously with 0.5 cc. of horse serum, and 5 died. Five guinea pigs were injected subcutaneously with 50 mg./kg. of procaine, and 30 minutes later injected with 1.0 mg./kg. of histamine into the penile vein. All animals died. Five control animals also died from single injections of 1.0 mg./kg. of histamine into the penile vein. Eleven rabbits were sensitized (Arthus Phenomenon) by weekly subcutaneous injection of sodium penicillin G and horse serum. Five of these were injected twice daily with 50 mg./kg. of procaine. There were no demonstrable differences in the local reactions resulting from injections of sodium penicillin G (8000 units in 1 cc. normal saline solution) or horse serum (1 cc. of a 10% dilution in normal saline) between procaine treated and untreated animals. Accordingly, procaine failed to show protective effects against histamine toxicity, and such allergic manifestations as anaphylactic shock and the Arthus phenomenon. Thus the relief obtained with procaine in certain allergic states appears to be subjective, similarly to benadryl and pyribenzamine (Dreisbach: J. Allergy, in press).

Relation of meat intoxication in Eck-fistula dogs to hepatic dysfunction. VICTOR A. DRILL, *Department of Pharmacology, Yale University School of Medicine, New Haven, Conn.* An Eck-fistula was produced in three dogs who were then maintained on a high carbohydrate diet consisting of bread and milk with added iron salts, yeast and fat-soluble vitamins. The production of liver damage was followed by the bromsulphalein test. Following a suitable control period the dogs were fed only ground beef heart to produce meat intoxication. As the intoxication developed no increased impairment of liver function was observed, as measured by the bromsulphalein and alkaline serum phosphatase tests. This was observed even in two dogs that died during the meat intoxication. Two dogs with complete occlusion of the portal vein (occluded in three stages) and two unoperated dogs were similarly fed the beef heart diet but did not develop meat intoxication and also did not show evidence of impaired hepatic function during this period.

Antidiuretic activity of liver extracts and of urine from patients with hepatic cirrhosis. VICTOR A. DRILL and B. FRAME (by invitation). *Department of Pharmacology, Yale University School of Medicine, New Haven, Conn.* The concentrated urine from patients with hepatic cirrhosis and ascites showed antidiuretic activity when tested on hydrated rats, thus confirming the results of Ralli and associates. Dialysed antidiuretic urine lost activity much more slowly than did dialysed pituitrin. The active urines were also without oxytocic activity when tested on the guinea pig uterus. It is not definitely known at present if such antidiuretic urines derive their potency from the posterior pituitary gland. Such a substance may fail to be destroyed by an injured liver. However, the liver must also be considered as a possible source of such antidiuretic activity. When liver extracts were tested it was observed that crude liver extract, purified liver extract (15 U.S.P. units/cc.), and Intraheptol all had a definite antidiuretic effect. This effect of liver extract was produced by doses

as low as 0.1 cc. per 100 grams of body weight, administered intraperitoneally.

Toxicity and mechanism of action of p-nitrophenyl-diethyl-thionophosphate (E605). KENNETH P. DUBOIS, JOHN DOULL (by invitation), and JULIUS M. COON. *University of Chicago Toxicity Laboratory and the Department of Pharmacology, University of Chicago.* Lethal doses (20 mg./kg.) of E605 (Victor Chemical Works) injected intraperitoneally in cats elicited symptoms of generalized parasympathetic and central stimulation. Onset of symptoms was delayed and death occurred in about 1 hour with primary respiratory paralysis. Autopsy showed an intensely contracted intestinal tract and urinary bladder, and fluid in the respiratory tract. Sublethal doses (5 mg./kg.) produced tremors, muscular twitching, and fluid in the respiratory tract, with recovery in about 18 hours. A 90% propylene glycol-10% ethanol solution of the drug (2%) placed in the eye of the cat caused a moderate miosis with delayed onset. Atropine increased the resistance to the drug. In the phenobarbital anesthetized dog 2 mg./kg. intravenously gave a 25-fold augmentation of the depressor response to acetyl choline. In tests on rat brain cholinesterase *in vitro* 50% inhibition of activity was obtained by 1.5×10^{-6} M. The approximate LD-50 of E605 given intraperitoneally to rats was 6 mg./kg. After 20 mg./kg. 93% inhibition of brain and 78% inhibition of submaxillary cholinesterase activity occurred with an increase of free acetyl choline of the brain from 0.86 micrograms/gm. to 3.42 micrograms/gm. To ascertain whether cholinesterase inhibition by E605 is reversible rats were given 5 mg./kg. and groups containing at least 5 rats each were sacrificed at various times. The average per cent inhibition of cholinesterase was 94% at 0.5 hours, and in surviving animals was 89% at 1 hour, 46% at 2 hours, and 3% at 4 hours. These experiments demonstrate a reversible cholinergic action by E605.

The effect of methadon on the cholinesterase of the brain. G. S. EADIE, F. BERNHEIM, and D. B. FITZGERALD (by invitation). *Department of Physiology and Pharmacology, Duke University School of Medicine, Durham, North Carolina.* According to Scott et al. (J. Pharmacol. **91:** 147, 1947) the effects of methadon on the intestine and heart resemble those of morphine and demerol. The last two drugs inhibit the cholinesterase and the effect of methadon was, therefore, tested. Rat brains were ground in a blendor and washed five times in a large volume of acidified water. The preparation from one rat brain was suspended in 25 cc. of water and 4.0 cc. of this suspension was used in a total volume of 30 cc. The rate of acetylcholine hydrolysis by the enzyme was measured by electrometric titration in the presence and absence of methadon. Methadon definitely inhibits the esterase, and a provisional estimate of the dissociation constant methadon-esterase indicated that it is of approximately the same order of magnitude as the acetylcholine-esterase constant.

Enzymic hydrolysis of morphine esters. SYDNEY ELLIS. *Department of Physiology and Pharmacology, Duke University School of Medicine, Durham, North Carolina.* Atropine, monoacetylmorphine and benzoylcholine are hydrolyzed rapidly by the plasmas of some rabbits, whereas plasmas of other rabbits have little or none of these three activities. It has been shown that atropine esterase is an enzyme distinct from benzoylcholine esterase. It now appears that monoacetylmorphine is hydrolyzed by an enzyme other than those which attack atropine and benzoylcholine. Plasmas of rat (C. I. Wright: J. Pharmacol. **75:** 328, 1942), human, dog, sheep, and guinea pig have negligible activity on monoacetylmorphine. This ester is hydrolyzed by guinea pig liver. Wright found that human and rat livers can attack this compound. The enzyme of rabbit blood is found in the plasma and not in the red cell.

Differences in sensitivities of liver enzymes of rabbits and guinea pigs to heat, acetone drying and inhibition by physostigmine and neostigmine indicate that monoacetylmorphine is attacked by an enzyme distinct from benzoylcholine or tropine esterases. The enzyme which hydrolyzes the alcoholic acetyl group of monoacetylmorphine is able to remove the phenolic acetyl group of the diacetyl compound. However, the enzyme which attacks the latter group is unable to hydrolyze the former. Neither activity is related to acetylsalicylic acid hydrolysis. Specificity for phenolic or alcoholic acetyl groups is not found when phenylacetate and cyclohexylacetate are the subtrates. Monoacetylmorphine hydrolysis is not related to methylbutyrate hydrolysis but may be accomplished by the plasma enzyme which attacks tributyrin.

The therapeutic significance of detectable penicillin levels. N. ERCOLI, M. N. LEWIS (by invitation), B. S. SCHWARTZ (by invitation), M. H. WHITEHEAD (by invitation). *Dept. Pharmacology and Chemotherapy, Warner Inst. Therap. Research, New York City.* The duration of therapeutic activity of penicillin is regarded as depending on its presence in antibacterial concentrations in the blood stream. Under the artificial conditions of the laboratory test, the minimum amount which can be detected is 0.015–0.03 units/cc. serum; consequently, this concentration is accepted generally as corresponding to that of therapeutic effectiveness. We determined the relation between blood level and therapeutic activity in our study of a new preparation with prolonged duration, which consists of a suspension of crystalline potassium penicillin in oil with vasoconstrictor. First, we determined accurately the duration of the blood level in rats treated with varying doses of this preparation; then, we infected the animals at different intervals

when the blood levels were already *negative* (20–22 hours after treatment). We found that it is possible to protect rats against 100 lethal doses of Pneumococcus Type I, at time periods when the blood no longer contains detectable amounts of penicillin. These experiments indicate that penicillin might be therapeutically effective in serum concentrations below those which can be detected by the bacteriological methods in use, and—more generally—that these *in vitro* methods, though important for comparative purposes, are not suitable for the determination of the absolute value of the therapeutically active blood concentrations. Moreover, we found that with the preparation used the duration of *detectable* amounts of penicillin in the organs is more prolonged than in the blood. The prolongation of the penicillin tissue levels depends on the vehicle used for injection. For instance, a single treatment with the oily suspension with vasoconstrictor gives durations of tissue levels which cannot be obtained by repeated administration of multiples of the same dose injected in saline. Thus, blood levels and tissue levels are not necessarily parallel when different vehicles are used for injection. It might be assumed that a more prolonged duration of penicillin in the organs will produce relatively longer periods of therapeutic effectiveness.

Comparison of depth of local anesthesia obtained with 2% and 4% procaine solutions. FRANK G. EVERETT (introduced by NORMAN DAVID). *Department of Pharmacology, University of Oregon Medical School, Portland, Oregon.* Clinical experience indicates that for certain dental procedures 4% procaine solutions have advantages over the recommended 2% solutions for infiltration anesthesia. Such advantages have been difficult to measure by previous laboratory technics which are concerned primarily with onset and duration of local anesthesia rather than with intensity or depth. We have compared local anesthetic potency in two homologous teeth of the upper arch of dogs lightly anesthetized with sodium pentobarbital. Shallow occlusal cavities were prepared in each tooth to such a depth that equal minimal responses were noted when their sensitivity was tested by a weak faradic current of an inductorium. Simultaneous stimulation of each cavity revealed that after the first 10 minutes following nerve block with 4% procaine, responses were consistently absent or reduced to a stimulus which would produce a definite pain response in the contralateral tooth blocked with 2% procaine. This objective confirmation of the clinical advantages of 4% procaine solutions suggests the need for investigating the relative dangers of 2% and 4% procaine when so used. Such a study is now in progress.

Studies on the toxicity and metabolism of d-tubocurarine. G. M. EVERETT (introduced by R. K. RICHARDS), *Dept. of Pharmacology, Abbott Research Laboratories, North Chicago, Illinois.* There is considerable difference in the paralytic dose of d-tubocurarine for various species. The paralytic dose in mg./kg. for rabbits is 0.15, dogs 0.2, cats 0.2, mice 0.125, rats 0.075, guinea pigs 0.035. Early paralysis of the respiratory muscles and the development of anoxic jerks and convulsions are observed most often in mice, rats and guinea pigs. Artificial respiration with added oxygen aborts these anoxic effects. In anesthetized and unanesthetized cats and dogs just paralytic doses cause a transitory fall in blood pressure of 10 to 25 mm. Higher doses caused a marked vasodepression of 40 to 60 mg. with little recovery of the blood pressure. Vasopressor drugs are effective in restoring blood pressure to normal levels. No direct cardiotoxic effects were observed with doses up to 50 times the paralytic dose. Both the ECG and EEG remained essentially normal. There were no differences in the intravenous paralytic dose or duration of action observed between control, nephrectomized, hepatectomized (80%), or nephrectomized-hepatectomized rats. Studies in similarly operated rabbits showed no increase in sensitivity or duration of action of d-tubocurarine from normal controls. Upon giving a second dose after two hours to normal and operated animals no sign of residual curare was observed. These experiments indicate that the kidneys and liver are not essential for the metabolism of d-tubocurarine.

Some factors influencing the activity of cardiac glycosides in the rat. A. FARAH and E. SZMUSZKOVIEZ (introduced by Dr. JAMES M. DILLE). *Departments of Pharmacology, University of Washington Medical School, Seattle, Washington and the American University of Beirut, Beirut, Lebanon.* The toxic dose of g-Strophanthin is over a thousand times greater in the rat than in the cat. This is mainly due to the high elimination rate and the high resistance of the rat heart to the cardiac glycosides. The following factors concerned with the elimination and lethal dose of g-Strophanthin were studied: *Rate of administration:* A reduction in rate increases the lethal dose and experimental time. *Anesthesia:* Amytal and Urethan have been compared. With Urethan anesthesia the lethal dose is about seventy per cent lower than with amytal anesthesia. *Functional hepatectomy:* Markedly reduces the lethal dose and elimination of g-Strophanthin. *Liver damage:* Carbon tetrachloride in doses sufficient to produce liver damage reduces the lethal dose of g-Strophanthin. *Concentration of g-strophanthin:* If the rate of administration of g-Strophanthin is maintained at a constant value, the lethal dose is reduced by about 50% if the concentration of the glycoside infused is reduced from 5 to 2.5 mg. per cc.

Absorption and potency of small divided doses of dicumarol in man. DAVID W. FASSETT and E.

STERLING NICHOL (by invitation). *Cardiology Service, James M. Jackson Memorial Hospital, Miami, Fla.* During a recent study by Nichol and Fassett (Jour. Southern Med. Ass'n. **40**: 631, 1947) on the use of dicumarol over long periods of time to prevent recurrences of coronary thrombosis, considerable difficulty was encountered when attempts were made to estimate daily dosage requirements for periods of a week or two in advance. The available dosage forms of 50 or 100 mgm. capsules, while satisfactory for short term use of a few weeks, were entirely too cumbersome to allow a precise adjustment of daily dosage. In ten patients studied over periods varying from 8 to 44 months, the daily dose of dicumarol necessary to cause a reduction of plasma prothrombin content to about 20% of normal varied from 25 to 150 mgm. per day, and bore little relation to weight, diet, or other medications. By using 25 mgm. capsules given in divided doses throughout the day, it was found that the total daily requirement was the same or less than when the drug was given in single doses, and that in most cases there was less fluctuation in prothrombin level of the blood. It seems reasonable to assume that drugs such as dicumarol and propyl thiouracil which presumably interfere with normal chemical processes, should be given in such a manner as to be present at the site of action in fairly constant amounts.

Effect of various compounds containing iodine and other halogens on thiouracil goitre. J. K. W. FERGUSON and E. A. SELLERS. *Department of Pharmacology, University of Toronto.* Goitres were produced in rats weighing about 150 gms. by giving thiouracil (0.1%) in the drinking water. The compounds studied were either added to the drinking water or injected subcutaneously. After two weeks of treatment the rats were killed and the thyroid glands weighed. Thyroxine by injection prevented completely the enlargement of the thyroid gland by thiouracil. Sodium iodide in daily doses ranging from 0.03 mg. to 0.1 gm. decreased the enlargement by 20–30%. Iodoacetic acid (0.04–5 mg.) produced a similar effect. Diiodotyrosine by injection had no more activity than could be attributed to its content of iodine. Sodium chloride up to 0.1 gm./day had no effect, while sodium fluoride 0.6–18 mg./day increased appreciably the enlargement of the gland. An increase in size also occurred with sodium bromide (0.004–0.1 gm./day), although this was less marked than with fluoride. In spite of the effect of iodide on gland size, the decline in metabolic rates of animals receiving iodide with thiouracil did not differ from those of animals receiving thiouracil only. It is postulated that the action of iodides is antithyrotropic rather than by direct interference with the action of thiouracil. Studies are continuing on the effect of prolonged administration of iodides.

Effect of scilliroside on the respiration of cat heart muscle. MURRAY FINKELSTEIN (by invitation) and OSCAR BODANSKY. *Dept. of Pharmacology, Cornell University Medical College, New York City.* Scilliroside increased the rate of respiration of slices of cat heart muscle in the presence of glucose and in a reaction medium containing 18 mM phosphate and 0.8 mM calcium per liter. A concentration of 1.6 to 2.5×10^{-8} M caused a slow but continuing increase; two hours after the introduction of scilliroside, the rate had increased by about 20%. A concentration of 10^{-7} M scilliroside led to an increase of the same magnitude within the first hour; higher concentrations (10^{-6} to 5×10^{-6} M) led to maximal increases of about 40% within the first 30 minutes. In all these instances, the increases were sustained during the period of observation. In the absence of calcium, the introduction of scilliroside (1.6×10^{-6} M) caused a decrease of about 20% in the rate of respiration. In reaction media containing 0.8 or 1.6 mM calcium per liter but only 1.8 mM phosphate per liter, the rate of respiration was less than in the usual medium; the introduction of scilliroside caused marked increases of about 80 to 100%. In the absence of calcium and with low concentrations of phosphate (1.8 mM per liter) the rate of respiration was essentially normal; the introduction of scilliroside caused a decrease of about 40%.

Comparative tolerance of dogs, cats and rabbits to nicotine. J. K. FINNEGAN, P. S. LARSON and H. B. HAAG. *Department of Pharmacology, Medical College of Virginia, Richmond.* As part of a program concerned with determining the fate of nicotine in the body, we have compared the detoxification capacity for it on several species. Two series of determinations were made on barbitalized animals: (1) LD_{50} values as determined by instantaneous intravenous injection of the total dose and (2) the dose required to produce respiratory paralysis when given over an 8-hour period by continuous intravenous infusion. LD_{50} results were as follows: cats, 2 mg. per kg.; dogs, 5 mg. per kg.; rabbits, 9.5 mg. per kg. The results by the second series of determinations were: dogs, ca. 15 mg. per kg.; cats, ca. 22 mg. per kg.; rabbits, ca. 40 mg. per kg. Subtracting the values obtained in (1) from those obtained in (2), it is found that the capacity of these species to detoxify and excrete nicotine, in terms of mg. per kg. per 8 hours, is approximately as follows: dogs, 10; cats, 20; and rabbits, 30.

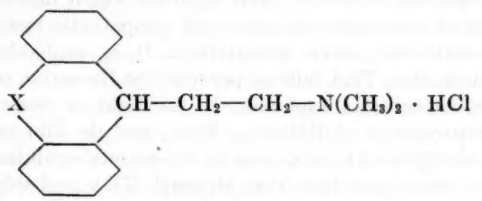
The chronic toxicity of thiourea. O. GARTH FITZHUGH, ARTHUR A. NELSON and OMA L. HOLLAND (by invitation). *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington 25, D. C.* Following the demonstration that orange decay was controlled by thiourea and that the juice of treated oranges contained thiourea (J. F. L. Childs and

E. A. Siegler, Ind. and Eng. Chem., **38:** 82, 1946), a 2-year chronic toxicity study was started. Albino rats, 21 days old, 18 to the group, were fed thiourea at levels of 1, 0.5, 0.25, 0.1, 0.05, 0.025 and 0.001% in a stock diet. All rats receiving 1, 0.5 and 0.25% thiourea died within the first year. Significant retardation in growth occurred at dosages of 0.025% or more. Dosages of 0.25% or more produced marked enlargement and, microscopically, marked hyperplasia of the thyroid. The 0.05 and 0.1% thiourea produced significant enlargement and hyperplasia of the thyroid. At all levels below 0.25% the production of liver tumors in the later months of the experiment was a more outstanding effect than were the thyroid changes. No tumorigenic effect of thiourea was seen at 0.25% or above, presumably because of the early death of all animals in these groups.

Statistical problems involved in the use of radioactive tracers in pharmacology. REX G. FLUHARTY (introduced by LLOYD C. MILLER). *Radioactivity Center, Massachusetts Institute of Technology.* The paper will first present a short discussion of the probability of radioactive disintegration and its effects upon the half life of the isotope, the statistics of the measurement, and the measurement sensitivity. An analysis will be made of some typical Geiger counter data. Limitations imposed by statistics will be pointed out, and criteria will be given for good operating conditions. Finally a summary will be given including the statistics of experimental factors such as sampling variations and limitations imposed by the techniques used.

Effects of certain "cardiac stimulant" drugs on coronary circulation and cardiac oxygen metabolism. ELWOOD L. FOLTZ, SAU KI WONG and JAMES E. ECKENHOFF (introduced by CARL F. SCHMIDT). *Department of Pharmacology, School of Medicine, University of Pennsylvania.* The effects of four "cardiac stimulant" drugs on the coronary circulation and the oxygen metabolism of the heart were studied in 16 experiments by a previously reported technique in intact dogs lightly anesthetized with pentobarbital sodium. Blood O_2 and CO_2 content was determined from samples obtained by catheterization of the coronary sinus, the pulmonary conus, and by direct puncture of the femoral artery. Respiratory studies of O_2 and CO_2 were accomplished on expired air collected in a Tissot spirometer. Cardiac output was estimated by the direct Fick method. Coronary flow was estimated by the N_2O technique as adapted to the coronary circulation (Am. J. Med. Sci., December 1947). After obtaining control values for cardiac output and coronary flow, one of the following drugs was injected intravenously and studied: theophylline with ethylene diamine, papaverine hydrochloride, nikethamide, and khellinin. Preliminary studies have shown that theophylline with ethylene diamine increased coronary flow in 2 of 3 experiments, but cardiac work and O_2 consumption was increased in all three. Papaverine decreased O_2 consumption in 4 experiments while coronary flow was increased only in the 2 experiments where a large dose (3 mgm. per K.) was injected; cardiac work was uniformly decreased. The response to nikethamide was variable. In 2 experiments with khellinin, no significant increase in coronary flow was observed.

The effectiveness of certain substituted ethylamines against histamine spray-induced fatality in the guinea pig. HOMER B. FREESE (by invitation), W. E. HAMBOURGER, and PATRICIA M. MICHIELS (by invitation). *From the Pharmacology Laboratories, Research Department, G. D. Searle & Co., Chicago, Ill.* Three series of compounds have been studied for antihistamine activity on guinea pigs using the histamine spray technique described by Loew, Kaiser, and Moore (J. Pharm. and Exp. Therap. **83:** 120, 1945). Animals were treated by intraperitoneal injection fifteen minutes before exposure to the histamine mist. Groups of untreated controls were run each day. The compounds studied have the following general fomulae:



Series I

X N—CH_2—CH_2—$N(CH_3)_2$ · HCl

Series II

In both series, X may be: oxygen, sulfur, methylene, C—C linkage (i.e., diphenylene), or by no linkage (diphenyl). The two series, therefore differ only in the methylene or nitrogen bond between the nucleus and the β-dimethylaminoethyl side chain. Series III consists of the 8-chlorotheophylline salts of the more active members of Series I and II. The MED is taken as the lowest dose which reduces the per cent mortality significantly when compared with the control mortality for the day. The MED of diphenhydramine HCl is arbitrarily assigned an activity index of 1, and on that basis indices are calculated for each of the compounds. The most active compounds are those in series I and II in which X is either sulfur or methylene, and their respective 8-chlorotheophylline salts in series III. The acridan compound (Series II, X = CH_2) is outstanding with an activity index of 16.

The effect of tetraethylammonium on the pressor response to anoxia and asphyxia. WALTER FREYBURGER (by invitation), LUIS R. CAPÓ (by invitation), and GORDON K. MOE. *From the Department of Pharmacology, the University of Michigan, Ann Arbor.* The pressor response to anoxia and asphyxia is thought to be due to central and reflex excitation of vasopressor mechanisms, including the secretion of epinephrine. Since the humoral mechanism of excitation of the adrenal medulla is cholinergic and analogous to the ganglionic synapse, and since TEA blocks the pressor action of high doses of acetylcholine, it was expected that this drug would prevent the hypertensive response by interrupting transmission between nerve ending and adrenal medullary cell. In the cat and rabbit, the pressor response was blocked by TEA, but in the dog the drug did not prevent, and often even potentiated, the hypertensive reaction. That some humoral pressor agent was indeed liberated in the dog in the presence of TEA was demonstrated, for vasoconstriction occurred in the denervated leg during exposure to asphyxia. That this humoral agent is epinephrine is likely, for Dibenamine, which converts the pressor action of epinephrine to a depressor action, also converts the pressor response to asphyxia to a depressor response, even in doses which fail to block completely the effects of sympathetic vasoconstrictor nerve stimulation. It is probable, then, that TEA fails to prevent the liberation of epinephrine in asphyxia. In the light of recent experiments of Bülbring, Burn, and de Elio on the effects of local anoxia on the adrenal medulla, we would conclude that although TEA probably prevents neurogenic stimulation of the gland, anoxia and asphyxia may produce in the dog a direct excitation which cannot be blocked by TEA.

Studies on ventricular fibrillation produced by epinephrine during hydrocarbon inhalation. SOLOMON GARB (by invitation) and MAYNARD B. CHENOWETH. *Dept. of Pharmacology, Cornell Univ. Medical College, New York, N. Y.* Experiments performed on 20 cats show that a sudden rise in systemic arterial pressure is not necessary for the production of ventricular fibrillation by epinephrine during hydrocarbon inhalation. Dibenamine protected 8 out of 10 cats from ventricular fibrillation in doses (3 mgm./kgm.) which did not reverse the pressor effect of epinephrine. Nor-epinephrine induced ventricular fibrillation during hydrocarbon inhalation in 5 out of 5 cats, while n-isopropyl epinephrine failed to induce it in 9 out of 9 cats, although in 4, the systemic arterial pressure was raised by aortic constriction. In 5 out of 5 isolated perfused cat hearts, ventricular fibrillation was produced by benzene and epinephrine. Experiments on the threshold of irritability of papillary muscles show that benzene and chloroform raise the threshold very markedly, while ethyl ether, ethyl alcohol and acetone raise it very slightly. Epinephrine produces spontaneous rhythms in those papillaries which have a small pacemaker nodule at the base. Analysis of 11 electrocardiograms taken at the onset of ventricular fibrillation reveals that there is no definite acceleration before the onset of fibrillation. A characteristic pattern was noted in each record, consisting of a small QRS followed by a large T wave, with ventricular fibrillation beginning at the peak of the T wave. These findings suggest that ventricular fibrillation is initiated when an area of myocardium containing a slightly higher concentration of hydrocarbon than the remainder of the heart fails to respond to a stimulus (QRS) but does respond to the following T wave.

Synergism in relation to toxicity of certain antiasthmatic drugs. R. A. GARDNER (by invitation), P. L. EWING, G. A. EMERSON and A. E. HANSEN. Death of an infant following the accidental consumption of what was believed to be sublethal amounts of the components of a proprietary preparation prompted the present study. Lethal levels of the component drugs, ephedrine, theophylline and phenobarbital, were quantified by intraperitoneal injections into a total of 507 healthy young adult white mice of a stock strain. The LD_{50} as estimated by the method of Miller and Tainter was found to be for ephedrine sulfate 307 ± 9.6 mg./kg., aminophylline 256 ± 3.5 mg./kg., and phenobarbital Na 192 ± 11.4 mg./kg. Inasmuch as it was not believed that the patient suffered from phenobarbital intoxication and since a few preliminary tests indicated a marked potentiation of ephedrine by aminophylline, combinations of varying fractions of the LD_{50} of each of these latter two drugs were tested. One-fourth of the LD_{50} of ephedrine plus 140 mg./kg. of aminophylline gave a mortality rate of 80%, indicating more than a four-fold increase in the toxicity of ephedrine by the concomitant action of sublethal doses of aminophylline, or a doubling of the toxicity of aminophylline by a sublethal dose of ephedrine. Phenobarbital decreases toxicity of both agents, alone or in combination.

Inhibitory effects of naphthoquinones and related compounds on glycolysis. CHALMERS L. GEMMILL. *Department of Pharmacology, University of Virginia, Medical School, Charlottesville, Virginia.* A study was made of the inhibitory action of some naphthoquinones and related compounds on the conversion of glycogen to lactic acid by extracts of frogs' muscles. The rate of glycolysis was determined by the liberation of carbon dioxide from a bicarbonate buffered solution in a Warburg vessel. It was found that the following compounds

partially or completely inhibited glycolysis in concentrations of 0.003 M or less: Sodium 1,2-naphthoquinone - 4 - sulfonate; 2 - methyl - 3 - bromo - 1,4 - naphthoquinone; 2 - chloro - 3 - (n) thiobutyl-1,4-naphthoquinone; hydroquinone; 2 - methyl - 3 - thioethyl - 1,4 - naphthoquinone; 2-methyl-1,4-naphthoquinone. The following compounds did not inhibit glycolysis in amounts of 0.003 M or less: Lawsone; Lapachol; Lomatiol; 1,4-naphthoquinone-2-thiopropionic acid; 2-methyl-3-isothiopropyl-1,4-naphthoquinone; 2,3-dimethyl-hydroquinone; 2,3,5-trimethyl-hydroquinone; 2-methyl-hydroquinone. It was necessary to dissolve some of these substances in small amounts of alcohol before adding to the enzyme mixture. Control experiments demonstrated that this amount of alcohol had no effect on the rate of glycolysis. The addition of cysteine reversed partially the inhibitory action of sodium 1,2-naphthoquinone-4-sulfonate.

Inhibitory effect of stilbamidine, guanidine and arginine on glycolysis. CHALMERS L. GEMMILL. *Department of Pharmacology, University of Virginia, Medical School, Charlottesville, Virginia.* Read by title. It was found that stilbamidine di-isothionate (Merck), guanidine hydrochloride (Eastman) and arginine monohydrochloride (Bios Laboratories) inhibited the breakdown of glycogen to lactic acid in extracts of frogs' muscle. The concentrations used were 0.003 M or less. Guanidine hydrochloride inhibited this reaction only when it was added before glycolysis was started. When it was added twenty minutes after the glycogen was placed in the reaction vessel, no inhibitory action was found. Other amidines and guanidines are now under investigation.

Malaria chemotherapy: 1. The response of trophozoite-induced infections with *Plasmodium cynomolgi* to various antimalarial drugs. CLARA S. GENTHER (by invitation), WANDA SQUIRES (by invitation), ROCHELLE FRADKIN (by invitation), and L. H. SCHMIDT. *Christ Hospital Institute of Medical Research, Cincinnati, Ohio.* This report is part of a study designed to determine whether infections with *Plasmodium cynomolgi* in the rhesus monkey can be utilized in the systematic search for chemotherapeutic agents which will cure human infections with *P. vivax*. This paper deals with the response of trophozoite-induced infections with *P. cynomolgi* to such drugs as quinine, quinacrine, chloroquine, oxychloroquine, and chlorguanide (Paludrine). More than 200 monkeys were infected with measured numbers of trophozoites. Treatment with one of the above drugs was initiated at various stages of the primary attack or during the first or second relapse in instances where the initial treatment was subcurative. The standard treatment period was 7 days. All drugs except quinine were administered once daily; quinine was administered both in single daily doses and at 6 hour intervals. Infections were considered cured when the blood remained free of parasites (by thick film examination) for 12 consecutive weeks after treatment, despite protein shock and splenectomy. Under the above conditions the daily doses (mgm. per kgm.) required for systematic cures were: quinine, 80; quinacrine, 10; oxychloroquine, 5; chloroquine, 2.5; chlorguanide, 0.6. Using quinine as the reference standard, the relative activities of the other drugs were: quinacrine, 8; oxychloroquine, 16; chloroquine, 32; chlorguanide, 128. These ratios compare with approximate values of 2, 4, 6, and 60 for the respective drugs in *P. vivax* infections. It may be concluded, therefore, that trophozoite-induced infections with *P. cynomolgi* and *P. vivax* are relatively similar in their response to the above drugs.

The efficacy of BAL (2,3-dimercaptopropanol) in the treatment of experimental lead poisoning in rabbits by FREDERICK G. GERMUTH, JR. (by invitation) and HARRY EAGLE. *From the Laboratory of Experimental Therapeutics of the U. S. Public Health Service and the Johns Hopkins School of Hygiene and Public Health, Baltimore 5, Maryland.* Acute and subacute lead poisoning were produced in rabbits by the administration of lead acetate. Five consecutive daily subcutaneous injections of lead acetate at a dosage of 240 mg. of compound per kg. per injection regularly caused the death of rabbits within 3 to 40 days. The intravenous injection of the same salt at a dosage of 12 mg./kg. per injection, repeated every 3 hours for 4 doses, produced an acute poisoning from which the animals died in 1 to 18 days. The intramuscular administration of BAL in peanut oil and benzyl benzoate, at individual dosages of 5 to 20 mg./kg., repeated every 4 hours, failed to protect these animals. In one group of animals with subacute (subcutaneous) lead poisoning, rabbits treated with BAL died significantly faster than did the corresponding untreated controls. The administration of BAL, however, caused a marked increase in the urinary excretion of lead. For 2 hours after a single injection of BAL at 20 mg./kg., the urinary excretion of lead increased 11- to 40-fold in animals with a subcutaneous depot, and 3- to 7-fold in animals injected intravenously. However, the magnitude of the excretion response diminished with each additional injection of BAL. Treatment with BAL had no demonstrable effect on the histopathological lesions produced by lead. It is believed that the failure of BAL to protect lead-poisoned animals may be in part due to the fact that it mobilizes

only a small fraction of the total body store of lead, and in part to the fact that the lead-BAL complex proved almost as toxic as the lead salt itself on intravenous injection.

Toxicity and cardiotonic activity of antibiotic lactones and synthetic analogs on the isolated frog heart. N. J. GIARMAN (introduced by E. L. MCCAWLEY). *From the Laboratories of Pharmacology and Toxicology, Yale University School of Medicine, New Haven, Connecticut.* A series of 30 lactones and synthetic analogs, some of which have been shown to possess striking antibacterial, antiprotozoal, and antiviral properties, have been studied to ascertain their cardiac toxicity and cardiotonic activity, known to be inherent in the lactone configuration. A special cannula introduced by Krayer as a modification of the Straub technique has been used in the entire investigation. The toxicity experiments revealed the important generalization that the unsaturated lactones were by far more cardiotoxic than either the saturated lactones or the furans tested. Three of the parent-structured unsaturated lactones were chosen for a more intensive study of the time for systolic standstill-concentration relationship with ouabain and digitoxin as points of reference. Thus if the minimum milli-molar toxic concentration of ouabain is arbitrarily designated as 1, that of 3-pentene-1,4-olide is 60, that of 2-pentene-1,4-olide is 600, that of the dilactone of pulvinic acid is 1, while that of digitoxin is 3. Several significant findings were disclosed by the experiments designed to show a cardiotonic effect on the frog heart made hypodynamic with low-calcium Ringers. All of the saturated lactones were devoid of activity except two: 2-keto-3-dimethylaminomethyl-butyrolactone and 2-keto - 3(3,3 - dimethylacryl) - butyrolactone, which bore the respective ratios of 1:1486 and 1:2570 in relation to the minimum milli-molar positive inotropic concentration of ouabain. The most effective unsaturated lactones were the dilactone of pulvinic acid (1:21), 3-phenyl-2-butene-1,4-olide (1:292), l-ascorbic acid (1:335), and 3-pentene-1,4-olide (1:600). The structurally related gamma-pyrones, kojic acid and meconic acid, yielded the positive inotropic ratios of 1:3400 and 1:327 respectively. The other cardiac glycosides used as reference agents were digitoxin and uzarin which showed the positive inotropic ratios of 1:2.4 and 1:7.0 respectively.

Distribution and excretion of benadryl (β-dimethylaminoethyl benzhydryl ether). A. J. GLAZKE, W. A. DILL (by invitation), and D. A. MCGINTY. *Research Laboratories, Parke, Davis and Co., Detroit, Michigan.* Benadryl concentrations were determined in the tissues of the rat and guinea pig at various time intervals after oral and parenteral administration. The drug was extracted from alkaline tissue homogenates with heptane, and determined chemically by a modification of the methyl orange procedure described by Brodie, Udenfriend and Dill (J. Biol. Chem., **168**: 335, 1947). Maximum concentrations of Benadryl were found 1 to 2 hours after subcutaneous administration. Guinea pig tissue showed about 2.5 times the concentration of Benadryl found in rat tissue. In both species, lung tissue showed by far the greatest concentration of Benadryl, followed in order of decreasing concentration by spleen, kidney, liver, brain, skin, muscle and blood.

Excretion of the drug and its degradation products was also investigated in rats, using radioactive Benadryl with carbon-14 in the alpha position of the benzhydryl group. About 50 per cent of the administered radioactivity was recovered in the first 24-hour urine specimens, principally in the form of a non-basic degradation product. The degradation process was also investigated *in vitro* with tissue slice preparations. A small amount of unaltered Benadryl was found to be excreted in human urine, using the Craig countercurrent distribution technic for extraction and identification of the compound.

The binding of barbiturates by human and bovine serum albumin. By LEO R. GOLDBAUM (by invitation) and PAUL K. SMITH. *From the Department of Pharmacology, The George Washington University School of Medicine, Washington, D. C.* Using an ultrafiltration technique described by Lavietes (J. Biol. Chem., **120**: 267–75, (1937) some of the factors affecting the binding of several barbiturates and thiobarbiturates by albumin have been investigated. The extent of binding varies with the pH, passing through a maximum at about pH 7.6–8.0 for all the drugs investigated. Increasing the albumin concentration from 0.5 percent to 5.0 percent increased the amount of pentothal bound from 30 percent to 85 percent. With increasing pentothal concentrations, the percent bound decreased from 90 percent at 50 mgm. per liter to 50 percent at 900 mgm. per liter. Thiobarbiturates are bound to a greater extent than their oxygen analogues (barbital 5 percent, thiobarbital 39 percent; pentobarbital 40 percent, pentothal 65 percent). At a constant pH of 7.3 and constant drug and phosphate buffer concentration, the percent bound usually increased with the length of the substituent groups (barbital 5 percent, ipral 13 percent, neonal 28 percent, pentobarbital and amytal 37 percent and ortal 65 percent). Those with a cyclic side chain were intermediate (phenobarbital 20 percent, phanodorn 25 percent, cyclopal 31 percent, evipal 29 percent). Allyl groups instead of ethyl groups increased binding slightly (dial 13 percent, alurate 20 percent, seconal 44 percent). Work is in progress

to determine if the extent of protein binding is associated with the relative concentrations in the plasma and tissues.

Studies on diffusion respiration. VII. Electrical cortical activity in dogs. ELI S. GOLDENSOHN (by invitation), EWALD W. BUSSE (by invitation), JOSEPH N. SPENCER (by invitation), WILLIAM B. DRAPER and RICHARD W. WHITEHEAD. *From the Department of Physiology and Pharmacology and the Department of Psychiatry, University of Colorado Medical Center.* Respiratory arrest was induced and maintained for forty-five minutes in ten dogs according to the technic of Draper and Whitehead (1944). E. E. G. records were examined before, during and for two months after the procedure. Pre-diffusion control and post-diffusion E. E. G.'s were taken under light thiopental sodium anesthesia. The pH of the superior vena caval blood averaged 7.32 at the onset of diffusion respiration and 6.69 after forty-five minutes of apnea. In the same period the blood sugar rose from an average of 91.9 mg. per cent to 150.3 mg. per cent. In the first few minutes of diffusion respiration the cortical waves decreased in amplitude but the frequency was not altered. In the next few minutes the cortical rhythm decreased in both amplitude and rate. Between five and fifteen minutes, intermittent periods of complete absence of electrical activity appeared. These periods lengthened progressively until at about fifteen minutes electrical activity could not be recorded even at the highest amplification. At this time the blood pH averaged 6.95. Complete absence of electrical activity persisted for the remainder of the period of apnea. On the resumption of respiration electrical cortical activity was seen in a few moments. This activity was slow and irregular for the first several hours. The E. E. G.'s appeared similar to the controls within two weeks after diffusion. The animals were sacrificed two months after the experiment. Pathological studies of the brain and other organs are being made.

Inhibitors of purified human plasma cholinesterase. AVRAM GOLDSTEIN (introduced by OTTO KRAYER). *Department of Pharmacology, Harvard Medical School.* An investigation into the inhibition of purified human plasma cholinesterase (fraction IV-6-3 of the system of Cohn et al.; J. Am. Chem. Soc., 68:459, 1946) by a variety of compounds belonging to widely differing chemical classes. A manometric method was used. It has been shown (Goldstein, A.: J. Gen. Physiol., 27: 529, 1944) that one molecule of physostigmine combines reversibly with one molecule of dog serum cholinesterase and that the inhibitor competes with acetylcholine for the active group of the enzyme. The compounds studied here can be similarly classified as reversible or irreversible. Reversible inhibitors can be further described as competitive or non-competitive with respect to acetylcholine. Mercuric and cupric ions inhibit irreversibly. Methylene blue, acriflavin, morphine, atropine, strychnine, amphetamine, phenobarbital, sulfanilamide, procaine, choline, acetyl-β-methylcholine inhibit reversibly *but non-competitively*. The 1:1 relationship of molecules inhibitor to enzyme active centers applies here. Since the interaction is reversible yet unaffected by the presence of substrate (acetylcholine), we assume the enzyme is inhibited by interaction at some point other than the usual substrate-combining center. Physostigmine, prostigmine and carbaminoylcholine inhibit reversibly and *competively*—ie., are displaced from combination by acetylcholine. We take this to indicate interaction at the substrate-combining center. These fundamental differences in mechanism of inhibition can not be correlated with the molar potencies of the inhibitors. Consequently molar potency (or dissociation constant) and molar combination ratio do not adequately describe an inhibitor. Further classification as *reversible* or *irreversible* and *competitive* or *non-competitive* facilitates a more rational approach to the problem of relationship between specific structural configuration and inhibition capability.

Chemotherapeutic studies on a series of dithiocarbamates and their bismuth derivatives. ANDRES GOTH and FABIAN J. ROBINSON (by invitation). *Dept. of Physiology and Pharmacology, Southwestern Medical College, Dallas, Texas.* One of us has reported that bismuth forms a highly active complex with "aspergillic acid". Further studies have shown that certain dithiocarbamates behave in a manner similar to "aspergillic acid" in being potentiated by bismuth. The dithiocarbamates studied were (in decreasing order of activity against Staph. aureus): Diethyl-, dimethyl-, morpholyl-, and isoamylethyl-. Sodium diethyl dithiocarbamate inhibited the growth of Staph. aureus in a dilution of 1:500,000. Some of the bismuth derivatives were much more active than the parent compounds. Bismuth diethyl dithiocarbamate inhibited the growth of Staph. aureus in a dilution of 1:10,000,000. Much of this activity was retained in 50% human plasma. The compound has a wide antibacterial spectrum, inhibiting M. tuberculosis and E. coli as well as Gram positive organisms. Inhibitory blood levels of 20–40 Staph. Units were obtained in two dogs which received 20 mg. per kg. of bismuth diethyl dithiocarbamate intravenously. Demonstrable blood levels persisted for 30 minutes. When bismuth thioglycollate and sodium diethyl dithiocarbamate were given by separate intramuscular injections to dogs inhibitory blood levels of 10–20 Staph. Units could be maintained for at least 2 hours. The

MLD of sodium diethyl dithiocarbamate for mice was found to be greater than 500 mg. per kg. intraperitoneally. Animal protection experiments are under consideration and will be reported.

Factors influencing the hypertensive action of desoxycorticosterone. D. M. GREEN and M. GLOVER (by invitation). *School of Medicine, University of Washington.* Single 20 mg. pellets of desoxycorticosterone were inserted subcutaneously in 30 young rats. Twenty-four additional animals received 10 pellets each. Equal numbers of males and females were used. Half the animals in each group were adrenalectomized. Blood pressure, weight and fluid intake were observed for 3 months. During the first 3 weeks all animals received isotonic saline to drink to minimize the post-operative mortality in the adrenalectomized animals. Thereafter, water was given to alternate animals. The immediate consequence of implantation was an abrupt rise in fluid intake, maximal within 10 days, followed secondarily by slow reversion toward control levels. Blood pressure showed no pronounced initial reaction but rose slowly during the three month period. In general, pressure elevation followed the initial rise in intake, and appeared inversely related to the secondary fall. At the 10 pellet dose level, major differences in degree of hypertension attributable to sex, salt intake or adrenalectomy were not demonstrated. At the 1 pellet level, maximum pressure in male animals definitely exceeded that in females, the majority of which failed to develop significant hypertension.

The effects of l-amino-l-phthalidylpropane hydrochloride on excised and intact intestine and uterus. CHARLES M. GRUBER and GOLDIE FREEDMAN KEYSER (by invitation). *Department of Pharmacology, Jefferson Medical College, Philadelphia, Pa.* In this study excised rabbit intestine was used as well as the intact intestine of non-anesthetized dogs with Thiry-Vella fistulae. Excised rabbit as well as intact rabbit, cat and dog uteri were also used. Racemate A and B of l-amino-l-phthalidylpropane hydrochloride decreased the general tonus of freshly excised rabbit intestine in 60 per cent of the experiments. The intestine kept in a refrigerator for 24 hours responded to the drug by a decrease in tone in 91 per cent of the experiments. An increase in the general tonus was noted in the remaining experiments. The effects of histamine acid phosphate, acetylcholine chloride and pilocarpine nitrate on the excised rabbit intestine could be antagonized by l-amino-l-phthalidylpropane hydrochloride. In five of the six non-anesthetized dogs with Thiry-Vella loops, the intravenous administration of l-amino-l-phthalidylpropane hydrochloride caused an increase in the general tonus of the gut. In the remaining animal 8 injections of the drug were followed in each instance by a decrease in the general tonus. The increased tonus caused by the drug can be antagonized by either barbiturates, or thiobarbiturates. Excised rabbit uterine segments respond to l-amino-l-phthalidylpropane by a sudden increase in the general tonus. Histamine acid phosphate, acetylcholine chloride or pituitrin, when added to the bath further increased the contraction of the uterus. The results on the intact uterus were variable. In dogs only contraction of the uterus was noted, but in cats and rabbits both decreases and increases in the general tonus were recorded.

The oxygen content of coronary venous blood as affected by anoxia and cytochrome C. JOSEPH H. HAFKENSCHIEL and JAMES E. ECKENHOFF (introduced by CARL F. SCHMIDT). *Department of Pharmacology and Harrison Department of Surgical Research, School of Medicine, University of Pennsylvania.* Utilizing the bubble flowmeter and the catheterization technic of investigating the coronary circulation, a study has been made of the normal oxygen content of coronary venous blood and of the effect upon it of anoxia and of intravenously injected cytochrome C. In the experiments where the bubble flowmeter was used, coronary venous blood was obtained from a cannulated great cardiac vein; where the catheterization technic was used, coronary venous blood was withdrawn from the coronary sinus. In 20 control experiments, coronary venous blood contained 3–6 volumes per cent of oxygen when arterial blood contained 16.5 to 19.0 vol. %. In a series of 5 other dogs, the average control oxygen content was: arterial 19.8 vol. % and coronary venous 6.5 vol. %. With the inhalation of 8% oxygen these values became 9.5 vol. % and 2.0 vol. %. Cytochrome C was injected intravenously, in amounts varying from 3.8 to 12.5 mgm./kg., into 8 dogs made anoxic by breathing 8 to 14 per cent oxygen mixtures. In 7 instances there was no evidence of an increased removal of oxygen from the coronary blood. In one dog, the A-V oxygen difference was increased but only because the arterial oxygen content was increased over the control values.

Toxicity of O,O-diethyl O-p-nitrophenyl thiophosphate (parathion). ERNEST C. HAGAN and GEOFFREY WOODARD (introduced by ARNOLD J. LEHMAN). *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington, D. C.* Increasing interest in the insecticidal properties of O, O-diethyl O-p-nitrophenyl thiophosphate (parathion) led us to study its action on warm-blooded animals. The oral LD50's of this compound administered in corn oil solution are: mice, 25 mg./kg.; guinea pigs, 32 mg./kg.; male rats, 30 mg./kg.; female rats, 3 mg./kg. This sex difference in rats was not

observed when the compound was administered intravenously. The effects of oral or parenteral administration were lacrimation, salivation, intestinal hypermotility, and generalized fibrillary tremors. Deaths usually occurred in 24 hours, and resulted from respiratory failure. Application of 0.5% concentrations in propylene glycol resulted in a miosis lasting over 8 hours which could be reversed within a few minutes by instillation of atropine. Prior administration of physostigmine (1 mg./kg. intraperitoneally) diminished the symptoms and increased the survival time of rats given oral LD50 doses of parathion. These observations suggest that parathion possesses anticholinesterase activity. Nicotine or atropine exerted some protective effects in animals poisoned by parathion. Daily administration of sublethal doses of parathion indicates cumulative effects.

Chloroquine in human giardiasis. ALVIN S. HAMBLY, JR. (introduced by Hamilton H. Anderson). *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco, California.* (read by title). Quinacrine [*3 - chloro - 7 - methooy - 9-(1 - methyl - 4-diethyl-amino)-acridine dihydrochloride*] has been used with some success in control of intestinal giardiasis (*G. lamblia*) in man. Since chloroquine [*7 - chloro 4(4 - diethylamino - 1 - methylbutyl-amino*) quinoline diphosphate], which is chemically related, has proved more active against malaria, trial in human giardiasis seemed indicated. Twenty-six patients who harbored *G. lamblia* were divided into two groups; 14 were given chloroquine and 12 were not-treated. The oral dose was 2.2 mgm. per kilo given daily for 7 days. Three of the 14 treated patients remained clear of infection during a 3-month follow-up period during which an average of 12 or more stool specimens were examined. Half of those treated were freed of diarrhea during or immediately after therapy. The untreated controls continued to harbor giardia throughout the follow-up period. Among treated individuals the drug caused no change in tests of urine, blood, liver, kidney or cardiac function, nor other objective evidence of intolerance. These observations do not confirm the report of Basnuevo and Sotolongo (Rev. de Med. Trop. y Parasit., Bact., Clin. y Lab., 12: 113, 1946) who "cured" 12 of 15 patients with giardiasis using chloroquine. The dose schedule of these workers was 1.0 gm. the first day, followed by 0.5 gm. for 2 to 4 days in adults.

Some extra-cardiac effects of digitalis. CARROLL A. HANDLEY and MARGUERITE LAFORGE (by invitation). *Dept. of Pharmacology, Baylor Univ. College of Medicine, Houston.* We have recently observed (Jour. Pharmacol., 89: 97, 1947) that the administration of non-toxic doses of digitalis to dogs results in prominent changes in the fluid compartments of the body. Further studies on the extra-cardiac effects of digitalis have revealed that a considerable elevation in blood potassium occurs concomitant with the shift in body fluids. The magnitude of these various effects were as follows: increase in "extracellular" fluid, 8-20%; decrease in plasma volume, 15-30% (checked by plasma protein determination); increase in blood potassium, 50-150%. The maximum changes occurred in 4-8 hrs. The potassium changes seemed to persist longer than the effects on body fluids.

Action of peroxides on endameba histolytica. EDER L. HANSEN and RACHAEL K. REED (introduced by Hamilton H. Anderson). *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco, California.* The requirement of *E. histolytica* for a high reduction potential in its environment is of interest and prompted the investigation of the action of peroxides as amebacidal agents. The following compounds were tested in liver-proteose-peptone medium (Parasitology, in press); 48 hours exposure at 37°C.

	Greatest dilution amebacidal
Superoxol	1/1000 by volume
Tertiary butyl hydroperoxide*	1/50,000
	Greatest concentration tested, no effect on ameba
Tetra ethyl phosphate†	1/10
Urea peroxide	1/2500
Sod. pyrophosphate peroxide	1/2500

* Fungicidal activity reported by Combes (N. Y. State J. Med., 37: 1927, 1937).

† Peroxide nature not definitely proved.

The amebacidal activity of tertiary butyl hydroperoxide is believed to be due to its stability which is greater than that of the other peroxides tested. This compound was given to naturally infected macaques at 100 mgm./kg. daily for 10 days in gelatine capsules orally. During an 11-16 day post-treatment period, the iron-hematoxylin stained cysts appeared unusually vacuolated, and the nuclei were fragmentary. Ten days later the amebae regained their normal appearance. Non-pathogenic amebae did not show morphologic changes. The monkeys exhibited no apparent toxicity, either local or systemic.

Functional analysis of the chronic neurotoxic action of streptomycin. JOSEPH E. HAWKINS, JR. and WALTER J. O'SHANNY (by invitation). *Merck Institute for Therapeutic Research, Rahway, New Jersey.* The earliest signs of cumulative intoxica-

tion in cats receiving repeated doses of streptomycin subcutaneously are copious salivation and slight ataxia. Salivation, which usually becomes established after several days, appears to involve a conditioned response. Ataxia, at first confined to the hindlimbs, increases in severity with continued dosing and is accompanied by a progressive loss of the nystagmus evoked by rotation, affecting first the post-rotational and later the per-rotational response (Fed. Proc., 6:125, 1947). Similar effects have been produced with all salts of streptomycin tested, but the time of onset may vary with the purity of the preparation, of which the microbiologically determined chemotherapeutic potency is not necessarily a complete measure. Thus when the hydrochloride (potency 668 units per mg.) was administered to 4 cats in daily doses of 100 mg. base per kg. body weight, 13 to 24 doses (mean 19) were given before symptoms appeared, while in 4 cats treated with equivalent doses of the crystalline calcium chloride complex (potency 685 units per mg.) 32 to 51 doses (mean 38) were required. When the drug is withdrawn, the ataxia and other manifestations of vestibular disturbance gradually disappear. After a few months only a slight awkwardness remains, but little or no recovery of nystagmus occurs in cases of severe loss. Comparison with the results of labyrinthectomy suggests that impairment of peripheral vestibular mechanisms alone is insufficient to account for the neurotoxic effects. Certain closely related central vestibular, cerebellar and optomotor functions appear also to be affected by streptomycin.

Pharmacological actions of O-O-diethyl O-p, nitrophenyl thiophosphate. LLOYD W. HAZLETON and EMILY GODFREY (by invitation). *Hazleton Laboratories, Falls Church, Virginia.* This compound is also known as Thiophos 3422 Insecticide. The most prominent actions of diethyl p-nitrophenyl thiophosphate are muscarinic, but these are not complete and there is evidence of nicotinic action as well. In rabbits dermal application produces diarrhea in lethal or nonlethal doses and there is some salivation. Myosis is not prominent following systemic administration, but application to the eye produces hyperemia and maximal constriction with return to normal overnight. This action is not reversed by atropine but may be prevented by prior administration. In the anesthetized dog intravenous administration of diethyl p-nitrophenyl thiophosphate is followed in eight to ten minutes by increased pulse pressure and decreased cardiac rate, accompanied by decreased respiratory volume with little change in rate. At the extreme, when respiration is limited to inspiratory spasm, both cardiac and respiratory effects may be almost completely relieved by 1 mg./kg. of atropine sulfate. Atropine given prior to oral administration of diethyl p-nitrophenyl thiophosphate exerts an appreciable life saving effect in rats and mice. Atropine does not antagonize the peripheral muscular spasms which appear to be nicotinic in nature.

The action of p-dimethylaminobenzenediazo sodium sulfonate on carbohydrate metabolism. ROY G. HERRMANN (by invitation) and KENNETH P. DUBOIS. *University of Chicago Toxicity Laboratory and the Department of Pharmacology, University of Chicago.* Read by title. p-Dimethylaminobenzenediazo sodium sulfonate (DAS) has been employed as a rodenticide in Germany. This use together with the chemical similarity of DAS to the carcinogenic azo dyes stimulated our interest in the mechanism of action of this compound. The approximate LD_{50} of DAS to rats was 15 mg./kg. After uniformly lethal doses symptoms appeared in about 2 hours and consisted of depression, flaccidity, and terminal convulsions. After 30 mg./kg. of DAS intraperitoneally to rats, which resulted in death in 3 to 8 hours, a rise in blood glucose from an average normal value of 94 mg.% to 172 mg.% occurred in 3 hours followed by a decrease to 20 mg. % at the time of death. Liver glycogen decreased from 2.14% to 0.114% in 5 hours after 30 mg./kg. of DAS. The administration of glucose prevented the terminal convulsions and insulin and adrenal-demedullation counteracted the hyperglycemia but did not prevent death. DAS inhibited the oxidation of glucose by rat liver slices *in vivo* and *in vitro*. A final concentration of 1×10^{-4} M DAS produced 53% inhibition of the oxidation of glucose by liver slices and at 3 hours after 30 mg./kg. of DAS a 38% decrease in the oxygen consumption of liver slices was noted. Enzyme studies have indicated that DAS inhibits an oxidative enzyme other than succinic dehydrogenase, cytochrome oxidase, or malic dehydrogenase since no decrease in the activity of these enzymes was observed in livers from rats poisoned by 30 mg./kg. of DAS.

The water-soluble B-glucoside of desoxycorticosterone acetate; its adrenal cortical activity in adrenalectomized dogs. E. HERROLD (by invitation), H. HAYS (by invitation), G. HOLMQUIST (by invitation), B. RICHARDS (by invitation), E. OPPENHEIMER. *Division of Pharmacology, Research Department, Ciba Pharmaceutical Products, Inc., Summit, New Jersey.* Initial experiments have confirmed Meier's work (Meier, R., Gysel, H. and Mueller, R., Schwz. med. Wschr. **74**: 93, 1944) on the doses of the B-glucoside of Desoxycorticosterone Acetate necessary: (1) to maintain an adrenalectomized dog; (2) to correct the syndrome of severe adrenal insufficiency. On a molar basis, 1.6 times as much glucoside as DCA in oil was required to maintain an adrenalectomized dog previously supported with DCA in oil; Meier reported that twice the dose of glucoside, on a molar basis, pro-

vided maintenance but that 1.5 times the dose of DCA in oil did not prevent slight symptoms of insufficiency. In our experiments, the dogs were first "normalized" after operation by minimal doses of DCA in oil. We found that 0.35 mg. of DCA glucoside/kg. of body weight maintained the animal; Meier recommends 0.4 to 0.6 mg./kg. Symptoms of severe adrenal insufficiency appeared within 24 to 72 hours after withdrawal of the glucoside and once initiated, progressed with unique speed. Meier has recommended from 16 to 50-fold the normal maintenance dose to correct severe adrenal insufficiency, and our experiments have confirmed this range. Because the water-soluble B-glucoside of DCA can be administered intravenously, its corrective action in severe adrenal insufficiency is rapid and dramatic.

The effects of 2,4 dinitrophenol and thiouracil on chronic methanol poisoning. CHARLES H. HINE and THOMAS NATHANIEL BURBRIDGE (by invitation). *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco.* Fifty rats of the Long-Evans strain were divided equally as to sex and weight (average 225 gm.) into 5 groups. The first group received methanol; the second, methanol plus dinitrophenol; the third, methanol plus thiouracil; fourth, dinitrophenol; and the fifth, thiouracil. These were given for the life of the animal up to 120 days. Seven per cent methanol, ingested as the only source of fluid, caused mortality in all animals over a period of 60 days. The mean survival time was 43 days and the mean weight loss was 99 grams. Simultaneous ingestion of 0.4% of 2,4 dinitrophenol increased the mean survival time significantly (76 days) in a second group of 10 rats but did not lower the mortality. Use of 0.2% thiouracil further increased the survival time (87 days) and decreased the mortality; 4 of 10 survived. The weight loss of the second group was greater and that of the third group less than noted in rats receiving methanol alone. Mean survival time was not affected (78 days) when 0.4% of 2,4 dinitrophenol was ingested alone (group four), but was increased (120 days–to end of observation) with 0.2% thiouracil (group five). There was a steady weight gain in group five for the first 100 days, after which weight loss occurred. All other groups showed an initial and progressive loss of weight. Bile staining of the liver, kidney, spleen, gut, and occasionally of the heart and lung was noted at autopsy in all rats receiving methanol (in the first three groups). Staining was most pronounced in those animals given methanol alone and least marked in those receiving thiouracil plus methanol. All groups which received methanol also showed an increased urine acidity (pH 4.0) and a slightly decreased visual perception of light.

The comparative activity of Some α substituted glycerol and glycidyl ethers. CHARLES H. HINE and HERBERT E. CHRISTENSEN (by invitation). *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco, California.* The comparative toxicity of a series of α substituted glycerol and glycidyl ethers has been determined in mice (30 animals for each agent) following oral and subcutaneous administration. These compounds have a curare-like action and produce loss of the righting reflex, muscular flaccidity and death through respiratory paralysis. The glycidyl compounds are generally more potent than their corresponding glycerol derivative. Replacement of the hydroxy groups of the β and γ carbons by one or more aliphatic groups decreased the potency. Delayed death occurred with most of the compounds following 24 hour exposure to the saturated vapors. Percutaneous absorption caused death with only the more toxic derivatives, such as glycidyl phenyl ether and glycidyl isopropyl ether. There was no irritating effect following topical application of the aliphatic compounds but the aromatic derivatives produced irritation and a leathery induration in the unabraded shaved skin of albino rabbits. The LD_{50} for both species was within the same range for any one of the compounds tested. Rabbits were slightly more resistant to oral administration. Neck drop was elicited in this species as an early sign of muscular paralysis after safe as well as fatal doses.

Certain aspects of the acute toxicity of beryllium following intraperitoneal injection. HAROLD C. HODGE (by invitation), ELLIOTT A. MAYNARD (by invitation), and WILLIAM L. DOWNS (by invitation). Introduced by HARVEY B. HAAG. Following intraperitoneal injection in rats and mice, the LD50 dose based on 24-hour mortalities was determined for various beryllium compounds. A few rabbits and guinea pigs were also injected. The soluble beryllium salts gave indication of toxicity of varying degree; the insoluble salts were practically non-toxic. Saline solutions were less toxic than aqueous solutions. Young (weanling) rats and mice were more resistant to beryllium toxicity than were older animals.

Two new analgesics. HELEN L. HOLLAND (by invitation and E. G. GROSS. *Department of Pharmacology, School of Medicine, Iowa City, Iowa.* The Cis (Nu 1196) and Trans (Nu 1779) forms of 1,3-Dimethyl-4-phenyl-4-propionoxy piperidine hydrochloride have been compared with morphine for analgesic potency in normal human subjects and in dogs, using the Wolff-Hardy technique. The analgesic potency of the Cis form in dogs is slightly greater than morphine, while the Trans

form is about 4 to 5× as potent as morphine. In man, the Cis form is somewhat less analgesic and the Trans form about equal in analgesia potency to morphine. The duration of analgesia of the Cis form is shorter and that of the Trans form about the same as morphine. Mild sedation is produced by the Cis form and rather marked sedation by the Trans form. Untoward effects of the Cis form are relatively few, only slight dizziness, minor itching and little or no nausea in doses up to 15 mgs. The Trans form produces marked dizziness, sweating, itching; nausea and vomiting are frequent at the 10 and 15 mg. level. No significant alterations in blood pressure or pulse rates have been observed with either form. No analgesic tolerance or alteration in R.B.C. or W.B.C. was observed in dogs receiving daily injections for 6 weeks.

A comparison of the toxicity of methadon and some related compounds. By JAMES O. HOPPE (by invitation) and LLOYD C. MILLER. *From Biology Division, Sterling-Winthrop Research Institute, Rensselaer, N. Y.* Acute toxicity studies in both mice and rats and subacute toxicity studies by subcutaneous administration to rats were made using aqueous solutions of the hydrochlorides of methadon, 6-dimethylamino-4,4-diphenyl-3-heptanone; its levo isomer; the *dl* form of a structural isomer, 6-dimethylamino-4,4-diphenyl-5-methyl-3-hexanone (Win 1783); ethyl-1-methyl-4-(m-hydroxyphenyl)-piperidine-4-carboxylate (Win 771); and 1-methyl-4-(3-hydroxyphenyl)-4-piperidyl ethyl ketone (Win 1539). The following acute LD_{50} values (mg. per kg.) for mice were found:

	Intravenous	Subcutaneous	Oral
methadon	13 ±1	18 ± 4	141 ± 6
levo-methadon	13 ±1	15 ± 3	111 ±12
Win 1783	22 ±2	40 ± 5	136 ± 8
Win 771	54 ±2	332 ±59	771 ±41
Win 1539	27 ±6	18 ± 5	120 ±23

The dose-mortality curves by subcutaneous administration were less steep than by either the oral or intravenous routes for each compound except Win 1539 which showed flat curves by all three routes. Thus doses of Win 1539 four times greater than those lethal to some mice failed to produce 100% mortality. Also this compound gave such erratic results by parenteral administration that the apparently greater toxicity by the subcutaneous route is insignificant. Subacute toxicity determinations were made in rats by daily subcutaneous administration for three weeks. The incidence of toxicological manifestations, particularly the inhibition of gain in body weight in the methadon series, was least with Win 1783 and greatest with *levo*-methadon. Win 1539 by this test was less toxic than *levo*-methadon, but more toxic than Win 1783.

Opposite effects in vitro and in vivo of a surface active agent on mycobacterium tuberculosis. S. H. HOPPER (by invitation), V. V. COLE, H. R. HULPIEU, and H. A. RAIDT (by invitation). *Depts. of Public Health, Pharmacology, and Microbiology, Indiana University School of Medicine, Indianapolis, Ind.* The agent used, Igepal, is described as a polyethylene alcohol condensate. The growth of Mycobacterium tuberculosis (H37 strain) was studied on Dubos' synthetic medium with and without albumin, and on beef infusion agar. When Igepal was added to the media there was complete inhibition of growth with concentrations of 0.005 per cent and above on Dubos' media and at 0.01 per cent and above on beef infusion agar. Guinea pigs were infected with human strains of tuberculosis and when more than half showed evidence of local involvement they were placed on the experimental diet. In a group of four receiving 0.5 per cent Igepal, all were dead 80 days later, whereas only one control was dead. The Igepal pigs all showed tuberculous involvement of the liver, while controls showed none. The spleens were larger in the Igepal pigs and showed a more widespread conglomerate type of tuberculosis. This experiment was repeated with a more virulent strain of the tubercle bacillus and 0.1 per cent Igepal in the diet. Eighty days after being put on the diet 3 of 13 controls and 9 of 12 Igepal pigs were dead. Organs involved were about the same in the two groups. There was no difference in spleen size but there was more of a conglomerate type of tuberculosis in the Igepal pigs. The results with 0.1 per cent Igepal were less marked than with 0.5 per cent but in the same direction. In vitro studies gave opposite results.

Procaine metabolism in dogs anesthetized by thiopental, ether and chloroform. H. R. HULPIEU, and V. V. COLE. *Dept. of Biochemistry and Pharmacology, Indiana University School of Medicine, Indianapolis, Ind.* Procaine HCl was given intravenously at various constant rates to dogs under light anesthesia (plane 1). Dogs under sodium thiopental withstood considerably larger doses and lived longer than those under chloroform. Dogs under chloroform were more resistant than those under ether. At death the blood levels of procaine were approximately the same under thiopental as under chloroform but under ether they were about 60 per cent lower. The brains of the dogs under thiopental had a higher concentration of both procaine and p-aminobenzoic acid than those under chloroform or ether. The livers from thiopental dogs had a lower concentration of procaine but a higher concentration of PABA than those from ether and chloroform dogs. The same concentration of procaine was found in the kidneys

of dogs under thiopental and chloroform but approximately 40 per cent less was found in the kidneys of these dogs given ether. PABA concentration in the kidneys was greatest under thiopental and least under ether. A time factor undoubtedly played a considerable part in the above distribution. Conversion of procaine to PABA was most rapid under thiopental and slowest under ether. For any given dose of procaine the ratio of blood PABA to that dose was largest under thiopental and smallest under ether. Under all three anesthetics the concentration of procaine was uniformly higher in the brains, livers and kidneys than in the blood. Concentration of PABA in livers and kidneys was higher than that in blood, while the concentration in the brains was relatively low.

Structural relationship to sympatholytic activity of certain B-chlorethyl amines. CARLTON C. HUNT (introduced by McKeen Cattell). *Department of Pharmacology, Cornell Univ. Medical College and Sloan-Kettering Institute, New York, N. Y.* B-chlorethyl amines of the following structure $R-N(R')-CH_2CH_2Cl$ were tested for a protective effect against lethal doses of epinephrine (LD 80-100) in mice. These compounds were injected subcutaneously (20 mg./kg.) 30 minutes prior to the intraperitoneal administration of epinephrine. Effective protective agents were injected in progressively smaller doses until 40% or more animals died after epinephrine. The following substitutions offered protection (minimum protective dose mg./kg. in parenthesis): R and R′ = benzyl (10,; p-methoxybenzyl (10); and m-methylbenzyl (5); R,R′ = benzimidazolemethyl, benzyl (10); a-naphthylmethyl, ethyl (0.1); a-naphthylmethyl, B-chlorethyl (0.5); benzyl, B-chlorethyl (0.5). Dimethyl B-chlor, B-phenylethyl amine was also effective (1 mgm./kgm.) whereas dibenzyl B-chlor, B-phenylethyl amine was inactive. The following substitution on both benzyl groups of dibenzyl B-chlorethyl amine (dibenamine) abolished protective activity: p-isopropyl, o-Cl, o-methyl. Substitution of p-nitro, o-Cl, and p-Cl on benzyl bis(B-chlorethyl) amine abolished activity. As previously reported substitution of the phenyl group directly to the N lacks activity. γ-phenylpropyl bis(B-chlorethyl) amine is inactive. Effective protective compounds were tested on the blood pressure response (cat) to injected epinephrine and acetylcholine (atropinized animal). In all cases the pressor responses were abolished. Benzyl bis(B-chlorethyl) amine and a-naphthylmethyl bis(B-chlorethyl) amine also prevented the fall in blood pressure following injection of epinephrine or isuprel. Sympatholytic activity appears to depend upon a phenyl or naphthyl group attached to N through 1 or 2 carbons, the N being attached to a B-chlorethyl and another alkyl or aryl group. As reported by Loew et al. a bi-phenoxyethyl derivative is also active.

Sudden deaths during chloroform and cyclopropane anaesthesia. DUNCAN E. HUTCHEON (introduced by G. H. W. LUCAS). *Department of Pharmacology, University of Toronto.* Guinea pigs, cats and dogs have been used to investigate the mechanisms causing sudden deaths under chloroform and cyclopropane. Changes in the cardiac rate and rhythm were observed during all stages of anaesthesia and after the injection of small doses of epinephrine. Under chloroform, 9 of 22 guinea pigs and 3 of 10 cats died abruptly as the result of ventricular fibrillation following the injection of epinephrine; the same dose of epinephrine produced no deaths in an equal number of animals. All cats except those that died of ventricular fibrillation developed a transient tachycardia following the epinephrine. Five of 10 dogs under chloroform and 3 of 7 dogs under cyclopropane died of epinephrine-induced ventricular fibrillation. Deaths occurred during light anaesthesia with chloroform and during deep anaesthesia with cyclopropane. The same dose of epinephrine causes a reflex bradycardia in the normal unanaesthetized dog and under ether, so that chloroform and cyclopropane alter the reaction of the heart to epinephrine. Chloroform and cyclopropane may increase the irritability of the myocardium or may decrease reflex vagal activity. In rats and rabbits ventricullar fibrillation could not be produced by epinephrine under either chloroform or cyclopropane.

The effect of curare on the esophagus of the dog. K. HWANG (by invitation) and K. R. UNNA. *Department of Pharmacology, University of Illinois College of Medicine, Chicago 12, Illinois.* The order in which individual muscles are affected by curare appears to be a function of the physiologic characteristics of the respective muscle. Reports in the literature regarding the effect of curare on the striated musculature of the esophagus are conflicting. The present work was designed to study the effect of d-tubocurarine on the cervical and thoracic portions of the esophagus which, in the dog, receive separate motor innervation (Hwang, Grossman and Ivy, Fed. Proc., 6:133, 1947). In dogs anesthetized with pentobarbital, contractions of the leg muscle and of the two portions of the esophagus were recorded. Their respective motor nerves were stimulated with condenser discharges of various frequencies for specified periods of time. Tetanic contractions of the esophagus were not sustained at stimulation rates of more than 50 per second. On the other hand, the leg muscle showed well sustained tetanus on stimulation at rates of more than 100 per second. Small amounts of

d-tubocurarine depressed the maximum rate of stimuli which were capable of causing sustained tetanus of the muscle. This effect occurred almost simultaneously in the leg muscle and in the different portions of the esophagus. D-tubocurarine paralyzed skeletal muscle and both portions of the esophagus before arresting the contractions of the diaphragm. Recovery of the diaphragm was followed by recovery of the intercostal and other muscles. However, the time for the full recovery of the esophagus was four or five times longer than that of the diaphragm.

The baljet reaction and pharmacodynamic studies of diginin and gitogenin. HARRY K. IWAMOTO and FREDERICK K. BELL (introduced by JOHN C. KRANTZ, JR.). *Department of Pharmacology, School of Medicine, Univ. of Maryland, Baltimore.* Recently a chemical method for the assay of digitalis preparations was described. This method utilizes the Baljet reaction in which a red-orange color is developed by the active glycosides which is proportional to their cardiotonic activity. Diginin, a glycoside isolated from *digitalis purpurea*, was found to exhibit a weak Baljet reaction and little cardiac action. Gitogenin, a sapogenin, was found to be inactive on the heart and gave a negative Baljet test. Since each of these substances is obtainable in significant yields along with the cardiotonic principles from *digitalis purpurea*, the importance of these properties with respect to the chemical assay is apparent.

Influence of some analgetics of obstetrical importance upon respiration of mature human placenta *in vitro*. HAL P. JAMES and ERNEST W. PAGE (introduced by C. H. HINE). *Divisions of Obstetrics and Gynecology and of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco* (read by title). Studies of the effects of various analgetic drugs upon the respiration of rat cerebral cortex slices have been reported by Fuhrman and Field (J. Pharmacol., **77**: 392, 1943) and by Elliott, Warrens and James (J. Pharmacol., **91**: 98, 1947). Inasmuch as the placenta is the only human tissue that may be conveniently studied *in vitro*, we wished to compare the influence of similar drugs upon this tissue. For this purpose, we have selected those agents commonly used for pain relief in labor. The influence of merperidine (demerol), methadon ("10820"), morphine, amytal and scopolamine upon the rate of oxygen consumption of placental tissue was studied by the direct method of Warburg. The fresh tissue was prepared in the cold by teasing the villi apart with blunt dissection. Approximately 200 mgm. samples were suspended in modified Krebs-glucose-ringer-phosphate solution. The oxygen uptake was determined at 20-minute intervals for 80 minutes before and after addition of the drug. The results are summarized below:

Drug	Concentration	Max. effect on QO_2	Time of max. effect
	moles/liter	*% of control value*	*minutes*
Demerol (merperidine)	0.00025	−10	20
	0.0005	−27	20
	0.001	−43	20
	0.0075	−83	40
Methadon (10820, amidone, dolophine)	0.00025	+22	60
	0.0005	+38	60
	0.001	+12	40
	0.002	−93	120
Morphine	0.00025	no effect	
	0.005		
Amytal	0.0001	+1	20
	0.0002	−9	60
	0.00045	−31	20
	0.0009	−63	80
	0.0018	−97	120
Scopolamine	0.00025	−11	20
	0.005	−21	20
	0.02	−56	20

Demerol, amytal and scopolamine appear to be true depressants, whereas morphine has little, if any, effect. Methadon stimulated respiration in low concentrations, but depressed it at higher levels. All concentrations used are considerably in excess of therapeutic, and the results do not correlate with effects of the same drugs upon the intact fetus. They do, however, correspond well with the above-mentioned studies on rat cerebral cortex.

Experimental bromism in man. E. M. JELLINEK. *Dept. of Applied Physiology, Yale University* (introduced by C. I. BLISS). In testing clinical reports on the occurrence of bromide psychosis at blood bromide levels ranging from 50 to 120 mg./100 cc., 78 psychologically normal persons and 60 psychotic patients were kept for several weeks at blood bromide levels around 200 mg./100 cc. Subjective and objective effects of the bromide administration were observed. A psychological test battery was employed to check on subjective reports. The statistical analysis takes account of the practice factors involved in these tests and attempts to arrive at the net effect of the drug. Even at fairly high bromide levels the normal subjects did not show gross psychological or neurological effects but only manifestations of sedation. The indications are that bromide psychosis at bromide levels reported in the clinical literature would occur largely in individuals with psychotic liabilities or at the onset of a psychosis.

Comparative analgesic and toxic effects of the optical isomers of methadon and isomethadon. ELIZABETH H. JENNEY (by invitation) and CARL C. PFEIFFER. *Department of Pharmacology, University of Illinois College of Medicine, Chicago 12.* Acute toxicity was determined by intraperitoneal administration of these drugs using the albino mouse (Harlan strain, 18–24 Grams, both sexes). With both drugs the d optical isomers were more convulsant and produced a less marked Straub-tail phenomenon. Analgesia was determined after subcutaneous administration in the guinea pig (method of Winder, Pfeiffer and Maison). An abnormally low therapeutic index was derived as a ratio of the mouse LD-50 to the minimal analgesic dose in the guinea pig. From these data dl Methadon has a slightly higher therapeutic index which is due entirely to a higher LD-50. Chronic toxicity studies in weanling rats (Sprague Dawley strain, both sexes) were made using a dose one-third that of the mouse LD-50. Injections were made subcutaneously daily for a period of five weeks. These data indicate that judged by the criteria of mortality, weight loss, and blood changes, d Methadon is the least chronically toxic and d Isomethadon is the most chronically toxic, while the two l Isomers are intermediate. The data further indicate the need for clinical studies on the pure l Isomethadon rather than the racemic mixture.

	Mouse LD-50*	S.E.	Slope	Minimal analg. dose*	Therap. index
dl Methadon	29	±2	11	12.5	2.3
dl Isomethadon	39	±3	9	12.5	3.1
l Methadon	11	±2	11	5	2.2
d Methadon	63	±3	14	20	3.2
l Isomethadon	25	±2	7	7.5	3.3
d Isomethadon	73	±6	9	30	2.4

*All doses are in mgm./kgm.

The relationship of cholinesterase inhibiting activity to the acute toxicity of some organic phosphorus compounds. H. WALTER JONES, JR. (by invitation), BERTRAM J. MEYER (by invitation) and LEONARD KAREL. *Toxicology Section, Medical Division, Army Chemical Center, Maryland.* The degree of *in vitro* inhibition of rat brain cholinesterase caused by 42 organic phosphorus compounds was compared with their acute (24 hour observation period) intraperitoneal toxicity to mice. The 38 compounds which proved to be weak inhibitors of cholinesterase (50% inhibition caused by molar concentrations greater than 2.5×10^{-4}) were also comparatively non-toxic (LD_{50}'s greater than 425 micromols/kg. or 100 mg./kg.), and no correlation between cholinesterase inhibition and toxicity was observed in this group. The remaining four compounds were potent inhibitors of cholinesterase. These were (1) tetraethyl pyrophosphate, (2) di-isopropyl fluorophosphate, (3) di-isopropyl chlorophosphate, and (4) diethyl p-chlorobenzene phosphonate. Two other potent cholinesterase inhibitors (A) and (B) (commercial confidential) were also investigated. The concentrations in which these six compounds caused 50% inhibition of cholinesterase activity and their LD_{50}'s in micromols/kg. and mg./kg. are presented in tabular form below.

Cmpd.	M Conc. for 50% inhib.	LD_{50}	LD_{50}
		micro M/kg.	*mg./kg.*
1	6.3×10^{-9}	4.00	1.16
A	1.0×10^{-8}	9.48	3.49
2	4.3×10^{-7}	47.5	8.74
B	4.2×10^{-7}	58.8	10.4
3	2.0×10^{-6}	146	29.2
4	1.6×10^{-5}	1120	300

When the logs of these LD_{50}'s in micromols were plotted aginst the negative logs of the molar concentrations causing 50% inhibition, a straight line was obtained. Thus the toxicity of these compounds appears to be a function of their potency as cholinesterase inhibitors.

Action of central nervous system depressants at different growth periods of salamander (Amblystoma punctatum) larvae. ALEXANDER G. KARCZMAR (by invitation) and THEODORE KOPPANYI. *Department of Pharmacology and Materia Medica, Georgetown University School of Medicine.* During the course of studies on the correlation of drug action and differentiation of the nervous system of Amblystoma punctatum, eight depressants were employed. Larvae of different ages (from 10 to 33 mm. in length) were immersed in standard concentrations of each of the depressants, and the anesthetic induction period (time in minutes elapsing between the immersion of larvae in solution and the onset of full anesthesia) recorded and plotted against larval lengths. For paraldehyde (1:200), ethyl alcohol (1:33), and tricain (MS-222; 1:7,500) the induction period was shortened gradually from a value of 40 to 30 to about 10 minutes, with a possible break in the curve occurring at larval length of 24 mm. It decreased abruptly for sodium barbital (1:20), nembutal (1:250), chloral hydrate (1:250) and chloretone (1:4,000), 12 mm. long larvae requiring more than two hours for development of full anesthesia, while 33 mm. long larvae were narcotized within from 5 to 10 minutes by chloretone and chloral hydrate and within 30 minutes by the two barbiturates. A possible break in the curve occurred at an early growth stage for chloretone, and at an

intermediate stage for the barbiturates and chloral hydrate. Finally, a 1:1,000 solution of acetanilid anesthetized the larvae within 6 to 15 minutes irrespective of larval length. Also, the minimal effective concentration of acetanilid (1:1,500) did not vary within the range of the larval age investigated, while the minimal effective concentrations of other depressants studied became progressively lower as the larval growth proceeded.

A potential rodenticide—"Fanyline". LEONARD KAREL. *Toxicology Section, Medical Division, Army Chemical Center, Maryland.* Subsequent to the report of Kalmbach in 1946 on the effectiveness of sodium fluoroacetate, "1080", as a rodenticide, attempts were made to find an equally effective verminicide which was, unlike "1080", relatively water insoluble. Potentially such a compound has been found in fluoroacetphenylhydrazide, hereafter called "Fanyline". Oral toxicity was determined in non-fasted animals by administering suspensions of "Fanyline" in 20% gum acacia through an oesophageal catheter or, in cats and dogs, by feeding capsules. Calculated median lethal doses were 9.1 mg./kg. for male rats, 44.9 mg./kg. for female mice, and 7.2 mg./kg. for pigeons; whereas the estimated LD_{50}'s were slightly less than 1.3 mg./kg. for rabbits, between 0.65 and 1.0 for guinea pigs, greater than 0.25 and less than 0.5 for cats, and between 0.1 and 0.25 for dogs. Mixtures of 1% and of 2% "Fanyline" in corn meal were highly acceptable to albino rats and mice, well-fed rats consuming poisoned meal even when presented to them side by side with their normal diet. In acceptability tests, all of 32 rats weighing 200 to 500 gms. and 16 rats weighing 65 to 82 gms. ate enough of the "Fanyline" to cause death. Furthermore, despite the relative nonlethality of the compound for mice, of 100 animals comprising four equal groups exposed to 2% poisoned meal, 33 consumed lethal quantities without pre-baiting, while of the 67 survivors, 55 consumed lethal doses after being pre-baited for one week. Toxicity and acceptability tests on wild Norway and Alexandrine rats are currently in progress in other laboratories.

Circulatory and adrenolytic actions of DHE and DHO. NORMAN W. KARR (introduced by NORMAN A. DAVID). *Department of Pharmacology, University of Oregon Medical School, Portland, Oregon.* Several hydrogenated ergot alkaloids have been shown to be less toxic and stimulant to smooth muscle than their naturally occurring precursors, while retaining full or augmented potency as adrenolytic agents (Rothlin, Bull. Schweiz. Akad. Med. Wiss., **2**: 249, 1947). We have compared dihydroergotamine (DHE), dihydrodergocornine (DHO) and ergotamine (Gynergen, Sandoz) with regard to reversal or diminution of pressor response to injected epinephrine, and alteration produced in the blood flow to a hind limb as measured with a bubble flowmeter (Soskin et al., Am. J. Physiol., **108**: 107, 1934). Dogs were anesthetized with intravenous sodium pentobarbital or with intragastric urethane and chlorobutanol. Under the conditions of our experiment, ergotamine has not consistently produced reversal or decrease in the pressor response to injected epinephrine, but in about 50% of the experiments a dose of 0.4 mg. per kg. has significantly diminished blood flow to the hind limb. This dose produces a marked rise in blood pressure, and frequently respiratory arrest and death. DHE and DHO in identical doses are well tolerated and without notable effects on blood pressure or respiration. Following either dihydrogenated alkaloid the pressor response to epinephrine is slight and transient and usually followed by a more marked and sustained fall in pressure. Neither DHE nor DHO has produced significant alterations in blood flow to the hind limb.

Toxicity of intravenous ammonium salts. NORMAN W. KARR and EDWARD L. HENDRICKS (introduced by NORMAN A. DAVID). *Department of Pharmacology, University of Oregon Medical School, Portland, Oregon.* Intravenous ammonium chloride has been reported to be a valuable treatment for alkalosis or certain types of oliguria, particularly where oral use of acidifying agents is precluded. While reactions characterized by pallor, sweating, and retching have been observed even with low concentrations (Schemm, reported before Western Soc. Clin. Res., Nov. 1947), search of the literature fails to reveal any systematic study of the toxicity of intravenous ammonium compounds. We have given ammonium chloride, carbonate, and bicarbonate to dogs by intravenous infusion, and find that all produce the same typical chain of toxic signs. The first is the appearance of occasional gasps, which become more frequent until the respiration consists only of a series of jerking gasps, with intervening periods of apnea. Simultaneously the heart rate becomes very slow, and often irregular. Fibrillary twitchings of the tongue appear, frequently followed by generalized fibrillations and tonic convulsions. If administration is continued, death soon ensues. This sequence of toxic signs is closely correlated with the rate of administration of the ammonium ion, regardless of the salt used. It has appeared with rates as low as 0.105 milliequivalents per kg. per minute, and a rate of 0.182 milliequivalents per kg. per minute has produced toxic signs in 15 minutes. Clinically ammonium chloride has been used in 2% solution, at the rate of 500 ml. per hour, which for a 70 kg. man, amounts to 0.045 milliequivalents per kg. per minute. Thus the safety margin for ammonium chloride given intravenously in this concentration is quite narrow,

ranging in 16 of our experiments from 2.29 to 4.37. This suggests the importance of close supervision of the rate of intravenous infusion of ammonium chloride, as double or treble the recommended rate might lead to serious toxic reactions.

Comparison of blood levels of thiopental (pentothal), 5-allyl-5-)1-methylbutyl)-2-thiobarbituric acid (surital) and thioethamyl in the dog. A. R. KELLY (by invitation), F. E. SHIDEMAN and B. J. ADAMS (by invitation). *Dept. of Pharmacology, Univ. of Michigan.* Using the Beckman spectrophotometer and a modification of the method of Jailer and Goldbaum, absorption and calibration curves and recoveries from plasma (96–100%) were obtained for three thiobarbiturates, thiopental, 5-allyl-5-(1-methylbutyl)-2-thiobarbituric acid (Surital), and Thioethamyl. Preliminary experiments demonstrated that withdrawal of blood from dogs in amounts up to 80 cc. did not significantly alter plasma blanks. It was also found that incubation of thiopental in plasma for 3 hours at 38°C. resulted in no appreciable diminution of recoverable drug. Blood level curves were obtained on dogs injected intravenously with sodium thiopental (20 mg./kg., 4% solution) and equimolar doses of Surital and Thioethamyl. The average duration of action and the blood level at the time of return of the righting reflex for each drug as follows: thiopental, 54 min., 1.2 mg. %; Surital, 94 min., 0.86 mg. %; Thioethamyl, 11 min., 2.1 mg. %. A comparison of potency based on the above blood levels and using thiopental as a standard of 100%, indicates that Surital has a potency of 139% and Thioethamyl, 57%. The shape and slope of the blood level curves for all three drugs was similar, indicating approximately equal rates of detoxication. A peak of approximately 3.5 mg. % was reached within one minute after completion of the injection, the level falling rapidly to 2.5 mg. % at 3–5 minutes. At 20 minutes it had reached 1.5 mg. % and leveled off so that at one hour the level was approximately 1 mg. % and at 4 hours 0.5 mg. %.

The anticurare activity of congo red and related Compounds. CHARLES J. KENSLER (introduced by MCKEEN CATTELL). *Department of Pharmacology, Cornell University Medical College.* Congo red and related compounds have been found to possess prophylactic and therapeutic anticurare activity in the frog when pure d-tubocurarine chloride (DTC) is the curarizing agent. Two pure erythrina alkaloids, beta-erythroidine (BE) and dihydro-beta-erythroidine (H_2BE) have also been studied. In contrast to the anti-DTC activity of congo red, no antagonism of the curarizing action of the erythrina alkaloids was observed. Diisopropyl fluorophosphate was less effective against DTC than congo red, but was as effective against BE as against DTC. Congo red is a moderately potent cholinesterase inhibitor in vitro but its failure to protect against curarization by the erythrina alkaloids as well as the lack of anti-cholinesterase activity of chlorazol fast pink (a related compound which possesses anti-DTC activity) indicates that the anti-DTC activity of congo red is not due chiefly to its anti-cholinesterase activity. Evidence has been obtained which supports the hypothesis that the anti-DTC activity of congo red and related compounds is due to a reaction occurring between DTC and the dyes. Congo red and related compounds have been found to prevent the dialysis of DTC through a cellophane membrane. No such effect was noted with BE and H_2BE. Congo red and other sulfonic acid compounds form a precipitate with DTC but not with BE and H_2BE. The DTC-congo red precipitate is soluble in excess congo red but not in excess DTC.

Methods for the evaluation of compounds with curare-like action. K. K. KIMURA (by invitation) and K. R. UNNA. *Department of Pharmacology, University of Illinois College of Medicine, Chicago 12, Illinois.* Curare-like compounds produce, on intralymphatic injection in frogs, progressive incoordination of spontaneous movements, loss of righting reflexes, and flaccid paralysis. By comparing the effects of indirect and direct stimulation of the muscle, the site of action of the drug can be verified with ease. Effects on other tissues, especially on the circulation, may be observed. Reversibility of the paralysis is a further indication of the selectivity of the action of the particular compound. Reproducibility of the effects of paralyzing agents on the muscles of the neck of the rabbit ("head-drop" method) depends on the speed of the intravenous injection and on the size of the animal. The effects are usually not recorded objectively. Marked relaxation of the abdominal muscles precedes that of the neck muscles. The narrow margin between the "head-drop" dose and the fatal dose too often results in the loss of animals. Intravenous injection in mice, likewise, produces head-drop. This method is more economical and allows the statistically valid determination of the "head-drop" dose on a uniform population. By determining the LD_{50} under identical conditions, the margin between paralyzing and lethal doses can be accurately assessed. Neither the rabbit nor the mouse "head-drop" method establishes the site of action of a drug. Potential curare-like compounds are, therefore, submitted to tests on frogs, before their effects are further studied on mammals.

The effect of pitressin on the hemodynamic responses of epinephrine and N-isopropyl-norepinephrine. THEODORE O. KING. *Department of Pharmacology, Georgetown University School of Medicine, Washington, D. C.* In a series of experiments on dogs, doses of epinephrine hydrochloride

(0.25 cc. and 0.5 cc. of a 1:100,000 solution) and of N-isopropyl-nor-epinephrine (0.25 cc. of a 1:100,000 solution) were administered by vein before and after the administration of pitressin. The pressor components of both drugs were potentiated by pitressin (see Table I), both as to height and duration. The vasodepressor effect of N-isopropyl-nor-epinephrine (# 1024) was usually abolished or

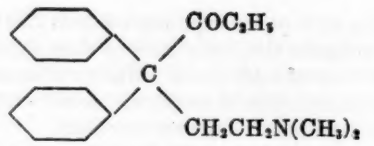
TABLE I

Total doses	Control	Pitressin (total)		
		0.5 unit	1 unit	2 units
Epinephrine HCl, 1:100,000				
0.25 cc.	+24 (11)	+32 (8)	+30 (7)	+36 (4)
0.50 cc.	+26 (8)	+34 (8)	+37 (8)	—
# 1024, 1:100,000				
0.25 cc.	−23 (10)	−2 (6)	+1 (7)	+12 (1)

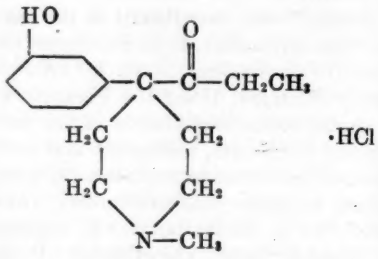
Ave. hemodynamic effects of epinephrine compounds before and after pitressin (nos. in parentheses = number of experiments). Plus indicates pressor, minus sign depressor effects.

at least reduced by pitressin. When the pitressin effects wore off, the injection of additional pitressin again produced enhancement of the effects of the epinephrine compounds. In those experiments where pitressin failed to affect potentiation of epinephrine B.P. elevations or reversals of the N-isopropyl-nor-epinephrine vasodepression, administrations of 0.1 mg. of erogotamine per Kg. produced potentiation and reversal respectively. It is suggested that the administration of a non-specific vaso-constrictor by opposing the vasodilator component of the above drugs enhances the pressor action of these agents and that the potentiation of epinephrine by cocaine (?), ephedrine or ergotamine is in part due to a non-specific vaso-constriction. Additional factors, of course, cannot be excluded. It may also be stated that in a statistically significant number of cases ergotamine reversed the depressor action of N-isopropyl-nor-epinephrine without accelerating the heart rate during the period of the actual rise of blood pressure.

Further studies on synthetic analgesics. A. C. KIRCHHOF. *The Division of Anesthesiology, University of Oregon Medical School, Portland, Oregon.* Two of the drugs reported by the United States Technical Intelligence Team as synthesized by the I. G. Farbenindustrie in addition to methadon have been given a limited clinical trial at the University of Oregon Hospitals. One of these, called by the Germans # 10582 (Formula I), was found to be rather inefficient in comparison with methadon. Analgesia was noted in 50 mgm. doses but side actions were too frequent to continue the study. The other, # 10720 (Formula II), called also Keto-Bemidone, in a very limited trial appeared to



I.



II.

merit further study since excellent analgesia was obtained with minimal side-actions.

Comparison of anesthetic action of acetanilid, tricaine (MS-222) and aliphatic depressants. THEODORE KOPPANYI and ALEXANDER G. KARCZMAR (by invitation). *Dept. of Pharmacology and Materia Medica, Georgetown University School of Medicine.* The kinetics of action of depressants depends, in Amblystoma punctatum larvae, on the anesthetic and on the larval stage. As to the latter, acetanilid (Fed. Proc., 6: p. 51), is a notable exception. Its speed of induction of anesthesia (see above, this issue) and its minimal effective concentration (1:1,500) are independent of the larval stage. Acetanilid action is reversible, the onset of action taking place within 5 to 15 minutes, and the recovery—from 2 to 3 minutes. Larvae, anesthetized with acetanilid, were subjected to multiple convulsive doses of metrazol, strychnine and prostigmine (o.c.). Convulsions were absent, but denarcotization failed to occur. Acetanilid also stopped convulsions already in evidence. Subanesthetic concentrations of acetanilid acted additively with subanesthetic concentrations of MS-222, nembutal and chloretone. Experiments in rabbits and dogs also indicate the C.N.S.-depressant action of acetanilid which is antagonized by metrazol; conversely, acetanilid prevents metrazol convulsions. Chloretone, tricaine, alcohol, and paraldehyde share with acetanilid the property of rapid reversibility of their action. On the other hand, the recovery time from chloral hydrate, nembutal and sodium barbital anesthesia is more prolonged. The ratio of recovery times for nembutal (1 hour) and for sodium barbital (4 hours) is comparable to that obtained in mammals. Experimental evidence shows that acetanilid is a general anesthetic requiring toxic doses for mammals but, for urodele larvae, concentrations below those of many aliphatic narcotics.

Anesthesia with Cyclobutane. JOHN C. KRANTZ, JR. and C. JELLEFF CARR. *Department of Pharmacology, Univ. of Maryland, School of Medicine, Baltimore.* The anesthetic properties of cyclobutane were studied in dogs and monkeys. The anesthetic syndrome produced by cyclobutane is comparable to that of cyclopropane. However, cyclobutane appears to be the more potent of the two gases. Blood pressure remains essentially unchanged, and the form and rhythm of the electrocardiogram remain normal under cyclobutane anesthesia. The automatic tissue of the heart is sensitized to epinephrine when dogs and monkeys are anesthetized with cyclobutane. The oil/water coefficient of cyclobutane is higher than that of cyclopropane, which correlates with the higher anesthetic potency.

Action of cysteine and of dimercaptopropanol in heartfailure caused by sodium bismuth tartrate. OTTO KRAYER and ALFRED FARAH. *Department of Pharmacology, Harvard Medical School.* In the isolated heart of the dog (heart-lung-preparation) sodium bismuth tartrate causes impairment of contractility and disturbance of rhythm. Hearts, in apparently similar condition of work capacity (as revealed by competence test) prior to the administration of the bismuth compound, vary greatly in sensitivity. Some hearts, especially those which have been exposed to other substances and have been in the experiment for sometime, may require as little as 25 mg. of sodium bismuth tartrate to produce severe failure; while in other hearts, especially in those freshly prepared, doses up to several grams may cause only moderate degrees of incompetence. Under certain conditions cysteine hydrochloride and dimercaptopropanol are able to restore normal function of the heart. The effect of cysteine is transient and the duration of action is proportional to the dose. If the first dose of the thiol compound is small in relation to the dose of sodium bismuth tartrate required to cause failure, no improvement occurs; rather, contractility is further impaired, or the heart is thrown into a state of severe arrhythmia or sudden stoppage of the heart is precipitated.

Vasomotor activity of NU-1683 and other drugs upon the rat meso-appendix. DONALD C. KUNZE (by invitation), J. RICHARD R. BOBB (by invitation) and HAROLD D. GREEN. *Department of Physiology and Pharmacology, Bowman Gray School of Medicine of Wake Forest College, Winston-Salem, North Carolina.* Various drugs have been tested on rats using essentially the technique of the Zweifach and Chambers rat meso-appendix test. The substances were tested as follows: Epinephrine was applied to the exposed mesentery in such dilution as to give a minimal constrictor response in the small muscular vessels. The drug to be tested was then injected intravenously. The reactivity to the topical epinephrine was again noted. When the test drug caused a higher concentration of epinephrine to be required, the drug was designated as a dilator, and when a lower concentration of epinephrine was required, the drug was considered to be constrictor. Epinephrine (0.3–0.7 microgram/100 gm.) had a constrictor action of slight degree (2×) for a short time (15 min.). Ephedrine (0.1 mgm/100 gm.) had a long-lasting (40 min.) constrictor action of small degree (2×). NU-1683(1-(m-hydroxyphenyl) -N^2-methyl-ethylene diamine dihydrochloride, an experimental sympathomimetic compound), (Hoffmann-La Roche, Inc.) (0.1 mgm–100 gm.) showed a constrictor action greater in degree than ephedrine (8×) but of the same duration. With this drug, the largest venules in the field became congested and occasionally the flow of blood completely stopped, to begin again after a short period of time. The entire bed showed a visible slowing of flow, even without the application of epinephrine, most marked on the venous side. Etamon (tetraethylammonium chloride) (2 mgm/100 gm.) showed a slight (2×), transient (10 min.) dilator action. Priscol (2 Benzyl 4,5 Imidazoline) (0.1 mgm/100 gm.) showed a dilator action of considerable strength (20×) for about 25 minutes.

The penetration of lead through the skin of the rat. FRIEDA M. KUNZE (by invitation) and EDWIN P. LAUG. *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington 25, D. C.* In the past experiments designed to demonstrate whether lead penetrates the skin have been equivocal. They were based chiefly on the attempt to show significant elevation of urinary lead following cutaneous exposure, but suffered either from lack of sensitivity or to technical difficulties in restricting lead intake to the cutaneous route. A new method (Laug, et al., J. Pharmacol. and Exper. Therap., **89**: 42, 1947) developed for the study of cutaneous penetration of mercury was applied to lead. This method utilizes the storage of lead in the kidney as a measure of the percutaneous penetration of lead. Rats were exposed to lead acetate, lead oleate, lead arsenate and lead tetraethyl either as aqueous or ointment preparations. From 77 to 150 mg. (on the basis of metallic lead) were inuncted into a 29 $(cm.)^2$ area of the skin of the back (8% of total body surface). The rats were wrapped in a special celluloid cuff which prevented oral contact. After 24 to 48 hours' exposure, the animals were sacrificed and the kidneys analyzed for lead. Small but significant penetration of lead followed exposure to the acetate, oleate and arsenate. Injury to the skin approximately tripled penetration. Markedly greater penetration occurred with lead tetraethyl,

the animals frequently dying within 24 hours from a total dose of 0.1 ml.

The daily dose of the mercurial diuretic and the maintenance dose to control congestive failure. NATHANIEL T. KWIT (by invitation), WALTER MODELL, LAWRENCE W. HANLON (by invitation), JOSEPH G. BENTON (by invitation), ELAINE W. COTLOVE (by invitation), MORRIS PEARLMUTTER (by invitation), SIDNEY M. GREENBERG (by invitation), MILTON L. KRAMER (by invitation), and HARRY GOLD. In the current methods for the treatment of congestive failure, the organic mercurial diuretics are used at infrequent intervals, rarely shorter than 3 or 4 days. Indications that mercuhydrin is excreted in less than 24 hours led us to its daily administration in a regimen of treatment of patients with congestive failure. The regimen consists of a diet containing 1 to 1.5 gm. of salt, 2 quarts of water, 1.2 mg. digitoxin at one time, followed by 0.2 mg. daily, and a daily intramuscular injection of 1 or 2 cc. of mercuhydrin. The results obtained with this regimen in 140 hospital admissions with advanced congestive failure were compared with those in 502 similar admissions in 4 large hospitals, treated by the methods in current use, in which, among other differences, the mercurial is given at infrequent intervals. The comparison showed that with the daily dose of the mercurial, congestive failure subsided completely in 90 instead of 50% of cases in an average period of 6 days instead of 15 days, the mortality being 9 in place of 23%. A chart of the daily weight shows when the patient attains the "cure" or "dry weight" (in 6 days). The "dry weight" is maintained by prolonging intervals in the maintenance plan, to prevent recurrences of failure, which constitute about 18% of hospital admissions for congestive failure by the current methods of treatment.

Pharmacological properties of trimeton, a new antihistaminic compound. ANNETTE LABELLE and RICHARD TISLOW (introduced by O. Krayer). *Biological Research Laboratories, Schering Corporation, Bloomfield, New Jersey.* Trimeton (γ - Phenyl - γ - (2 - pyridyl) - N,N - dimethylpropylamine) prepared by Drs. Sperber, Papa and Schwenk in the Chemical Research Division was studied for its potency, toxicity and other pharmacological properties. It prevented the lethal effect in guinea pigs of 1.1 mgm/kg. intravenous histamine dihydrochloride. The effective dose 50 per os was approximately 2 mgm/kg., and subcutaneously 0.1 mgm/kg. The protection afforded by 51 mgm/kg. orally lasted from 8 to 16 hours. 110 mgm/kg. orally, or 1.6 mgm/kg. subcutaneously protected guinea pigs for over 2 hours in a histamine aerosol mist in which the control animals died within 5 minutes. 24 mgm/kg. per os protected guinea pigs sensitized with species-different sera from anaphylactic death. Acute toxicity tests performed in mice, rats, guinea pigs and dogs by various routes compared favorably with other antihistaminic drugs in use. Examination of the blood and the organs of immature rats fed 25 mgm/kg. daily for 4 weeks did not reveal any difference between treated and control animals. In blood pressure experiments in dogs, Trimeton inhibited the depressor effect of histamine and potentiated the pressor effect of epinephrine. It had less spasmolytic activity than diphenhydramine hydrochloride as determined by the concentration effective in vitro in relaxing rabbits' intestine contracted with $BaCl_2$ or doryl. Trimeton did not have any antipyretic action as measured by a modification of the Barbour method. Solutions of Trimeton were less irritating to the rabbit eye and less anaesthetic to the guinea pig cornea than diphenhydramine hydrochloride.

Preliminary studies in the toxicity of beryllium: the effect of intratracheal injections in experimental animals. CHARLES W. LABELLE (by invitation) and MARTHA REID CUCCI (by invitation) (Introduced by Harvey B. Haag). With beryllium and its compounds are introduced into the lungs of rats via the trachea, the MLD for metallic dust and the insoluble compounds—oxide, carbonate, and carbide—are all greater than 200 mg of material per kilogram of body weight. The MLD of the soluble compounds, beryllium fluoride, sulfate, and oxyfluoride, was 15, 10, and 2 mg of salt per kilogram of body weight. Evidence is presented tending to show that the acute deaths result from a direct interference with lung function rather than a specific toxic effect of beryllium. The insoluble materials, while failing to show any distinct toxic effects of an acute nature, nevertheless lead to a more chronic type of damage resulting in a high rate of mortality after 100 to 250 days. Death is preceded by a series of inflammatory and necrotic changes in the lung, while changes in growth rate and increases in the polymorphonuclear leukocytes of the blood seem to offer the greatest aid in judging the progress of the reaction in intact animals. The pathological picture at death is not identical with that seen in man, and a series of concomitant exposures to debilitating agents such as mild infection, heat, and cold, has failed to alter the character or progress of the response.

Analysis of dust and fume hazards in a beryllium plant. SIDNEY LASKIN (by invitation), ROBERT A. N. TURNER (by invitation), and HERBERT E. STOKINGER (by invitation). (Introduced by Harvey B. Haag). A survey has been made of the industrial health conditions of a plant engaged in the manufacture of beryllium metal and alloys. The survey included an analysis of the dust and fume concentrations of the plant and a correlation

of these results with a past medical history of beryllium poisoning. The medical history of the plant from 1943 to 1946 showed 136 cases of toxicity attributed to beryllium compounds. Cases, for the most part, were confined to the preparation of the sulfate, the fluoride, the metal and the beryllium copper alloy. Typical symptoms are described. Dermatitis and skin ulcers accounted for 50% of all cases. Among the respiratory manifestations, chemical pneumonitis represented the severest form of the toxic symptoms; several deaths were reported in 1943. The general history showed a shift in case frequency away from the sulfate and beryllium alloy processes following improved workingconditions at these sites. At the beryllium fluoride and metal areas, frequencies increased, reaching the greatest proportions in 1946. The safety index ranged from 63.5 to 238.1 per million man hours of exposure. Acceptable normal plant practice is 4. The amount of atmospheric contaminants in selected areas was sampled for concentration and particle size, the selection being based on past medical history of hazard or on obvious contamination at the time of the survey. Sampling equipment consisted of a Modified Cascade Impactor, Filter Paper Dust Sampler, and a M.S.A. Midget Impinger. Near the fluoride furnace, fluoride concentrations—1000 fold that of beryllium—suggested a fluoride hazard in itself. Beryllium concentrations during operation averaged 0.065 m/mg^3 with a particle size of 2.46 microns (mass median). During operation of the beryllium metal furnace, beryllium concentrations varied from 1.4 to 4.7 mg/m^3. Fluoride samples were in the order of 1.7 mg/m^3 and the mass median particle size averaged 0.86 μ.

Near the rotary kiln drier the beryllium dust concentrations varied from 0.050 to 0.53 mg/m^3 fluoride concentrations were of the same order of magnitude. The particle sizes, however, were quite large, ranging up to mass medians of 10 micra.

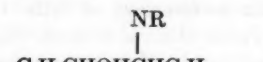
Several beryllium ores contained radioactive impurities. One lot was found to contain uranium concentrations up to 4% and thorium to 0.4%. The question of complicity of radioactive materials in beryllium toxicity is raised.

Tissue distribution of a toxicant following oral ingestion of benzene hexachloride by rats. EDWIN P. LAUG. *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington 25, D. C.* A biological assay method (Laug, J. Pharmacol. and Exper. Therap. **86**: 324, 1946) using the toxic response of the house fly has been applied to the determination of a toxicant in the tissues of rats exposed to gamma isomer of benzene hexachloride. Tests with the other isomers of benzene hexachloride, as well as the trichloro, heptachloro and octachloro derivatives, have shown that these compounds are only 1/100 as toxic as the gamma isomer. The latter is so toxic that 2 micrograms will kill 90 out of 100 flies within 24 hours. The toxic response to the gamma isomer is characteristic; when the insects are exposed to tissue extracts from animals poisoned with the gamma isomer their reaction is indistinguishable from that caused by the pure isomer. Therefore, it may be tentatively assumed that the toxicant in the tissues is the gamma isomer. Analyses of blood, liver, spleen, adrenal, muscle, brain, kidney and fat from rats which consumed diets containing 500 p.p.m. of the gamma isomer showed the presence of toxicant, the largest quantities (100 to 200 p.p.m.) occurring in kidney and fat. After one month feeding with benzene hexachloride the rats showed as much toxicant in their tissues as after four months. Detectable storage in liver, kidney and fat occurred even on diets containing 20 p.p.m. On the 500 p.p.m. diet approximately 1% of the injected benzene hexachloride appears as the toxicant in the urine, and only traces in the feces.

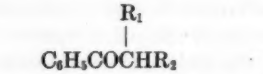
Molecular architecture of cells. C. D. LEAKE. *Univ. of Texas Medical Branch, Galveston.* Clark emphasized over a decade ago that the fundamental units in pharmacological activity are molecules of drugs and cells of living things. Drug action, however, is at a molecular level, and involves relations between molecules of the drug and molecules of the cell receptors. Thus, investigation of the molecular architecture of cells, including the organization of the various cell interfaces, becomes prerequisite to further clarification of the mechanism of drug action. Such study reveals the significance of short range (Van der Waals) polar association forces in drug-receptor relationships. The chief chemical factors involved are hydrocarbon, polar, and ionic groups, with spacing and stereochemical factors determining the resulting forces of association. These forces vary in geometrical and inverse proportion to the distances between relevant groups, suggesting the sensitiveness of the association forces to the orientation of the molecules concerned. Slight changes in spacing, dipole charge, or ionization may for or split an association complex, thus accounting for high specificity and rapid enzyme reversibility. Competition between related chemicals in regard to receptor complexes in cell interfaces or other molecularly organized cellular structures results in mass law equilibria. The approach to pharmacological problems through a study of molecular architecture of cells and receptor complexes permits an attempt to apply quantum theory to pharmacological mechanisms, as has already been shown to be so promising in connection with various mutafacient agents.

Studies on the relationship between chemical constitution and analgesic activity. ROBERT A. LEHMAN, HERBERT S. KUPPERMAN (by invitation)

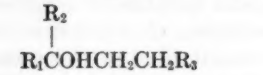
and JOY PHILLIPS (by invitation). *Department of Therapeutics, New York University College of Medicine, New York.* A program for testing synthetic organic compounds for analgesic activity has been set up using a modification of the rat tail method (See, Hardy, Wolff and Goodell, J. Clin. Invest., **19**, 649 (1940) and D'Amour and Smith, J. Pharmacol., **72**, 74 (1941)). The present technique consists of stimulation of the tip of the tail of the rat with a light beam of constant intensity and duration. It will be shown that under these conditions a linear relationship exists between the logarithm of the dose and the probit of the percentage of a group of animals failing to respond to the stimulus. The procedure used in rapidly screening a large number of compounds will be described in detail and results will be given for the several series listed below. From these data an attempt will be made to point out those structural features which appear to contribute to or are associated with analgesic activity.



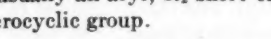
Where R is a short chain acyclic group. This series has been found to possess little or no activity.



Where R_1 and R_2 are usually heterocyclic groups.



Where R_1 is usually an aryl, R_2 short chain alkyl and R_3 a heterocyclic group.



Where R_1 is aryl or substituted aryl and R_2 is usually a heterocyclic group.

Where R_1 and R_2 include hydrogen, alkyl, aryl, aralkyl and heterocyclic groups. This series has been conspicuous in that marked activity is shown by many of its members.

Low toxicity and high therapeutic efficacy of combined sulfonamides. DAVID LEHR. *Dept. of Pharmacology, New York Medical College, Flower and Fifth Avenue Hospitals, New York City.* Several sulfonamides, even if closely related, could be dissolved simultaneously to the full extent of their separate saturation levels without the occurrence of precipitation. In the experimental animal the toxicity of the combinations sulfathiazole (ST)-sulfadiazine (SD), SD-sulfamerazine (SMD), SD-sulfapyridine (SP), SD-ST-SMD, SD-ST-SP, SD-SMD-sulfapyrazine, and SD-SMD-sulfacetimide, was found to be strikingly low as contrasted to the high mortality from comparable dosages of any *one* of the N_1-heterocyclic derivatives (D. Lehr, Brit. Med. J., in press). Combinations of three sulfonamides were significantly less toxic than combinations of two. Evidence derived from post mortem examinations and from chemical analyses of the blood, kidneys, and urine, proved that the low toxicity of sulfonamide combinations was due to the absence of crystalline deposition in the urinary tract (D. Lehr, Proc. Soc. Exper. Biol. & Med. **64**: 393, 1947). The antibacterial effect of combined sulfonamides, tested in vitro, was essentially additive. In some instances, combinations proved distinctly more active than one or several of their individual components at equal concentrations. More than 900 unselected patients of all age groups, with acute systemic infections were treated with equal partial amounts of SD and ST or SD and SMD, using the routine oral dosage. In infants and small children the latter combination was injected also subcutaneously. Therapeutic results were uniformly satisfactory and conspicuous in many instances because of the speed of improvement or cure. Effective blood levels of sulfonamide and high drug concentrations in the urine were readily maintained. Despite the intentional omission of alkalization, crystalluria was infrequent and no signs of renal irritation were encountered. Clinical studies with mixtures of *three* sulfonamides, as yet incomplete, indicate that the danger of concrement formation in the urinary tract can be entirely eliminated by combination therapy. Hency, mixtures of partial dosages should replace the use of single sulfonamides in systemic therapy, since mixtures combine a high bacteriostatic activity with a minimal renal toxicity.

Studies on the toxicity and pharmacology of some synthetic antispasmodics. DAVID LEHR. *Dept. of Pharmacology, New York Medical College, Flower and Fifth Avenue Hospitals, New York City.* The oral toxicity of di-(tributylethylamine)-β-naphthol-3,6-disulfonate (S_1), tributylacetamide (S_2), di-(tributylcarbinamine)-β-naphthol 3,6-disulfonate (S_6), tributyl ethyl nicotamide-(S_{51}), β-diethylaminoethyl-N-tributylcarbamate (S_{50}) and its hydrochloride (S_{60}), was studied in albino rats, partly also in mice and rabbits. The pharmacological action of these compounds with particular reference to antispasmodic activity was evaluated on the gut (rat, rabbit), on the uterus (rat, cat), on the rabbit eye, and the frog heart. The vascular response was tested by perfusion of the frog hind legs and the isolated rabbit ear, as well as by the effect on the blood pressure of intact cats and rabbits. Comparison was made with papaverine and atropine as standards for musculotropic and neurotropic spasmolysis, and in addition with

syntropan, trasentin, and profenil. S_{60} was found to be the most toxic compound, followed by S_{50} and S_6 which were about half as toxic, and S_1 which had one third the acute toxicity of S_{60}. S_2 and S_{51} were of such low toxicity that despite excessive dosages (15 and 10 gm./kg.), a M.L.D. could not be obtained. The pharmacological evaluation proved that all "S" compounds were essentially musculotropic antispasmodics. In the intensity of this effect S_{51} equalled papaverine, S_2 was twice as strong, S_1 and S_6 four times, and finally S_{50} and S_{60} about 5 times as strong. The latter two compounds produced irreversible changes on the isolated gut and frog heart. S_{51} caused slight stimulation of the frog heart and potentiation of acetylcholine action, whereas all other compounds induced only depression. S_2 and S_{51} effected a brief blood pressure rise and stimulation of respiration. The other 4 compounds lowered the blood pressure. None of these drugs had any action on the iris by local application, but all caused irritation of the conjunctiva which was least marked from S_{51}.

Influence of amidone, morphine and strychine on the Straub-tail-test of white mice. ALFRED LEIMDORFER. *Department of Pharmacology, University of Illinois, College of Medicine, Chicago.* After the intraperitoneal injection of 0.1 cc. of amidone-HCl (1% solution), but regularly after the injection of 0.2 cc., the tail was characteristically elevated. This phenomenon was more quickly produced and more pronounced by soft stroking along the spinal column in the thoracic region after the injection. Similar responses were observed after the subcutaneous injection of 1 mgm. morphine sulfate or after the subcutaneous injection of 0.15 cc. strychnine sulfate (1:1000). In further experiments it was found that strong pressure applied to the lumbo-dorsal region of the spinal column might elicit elevation of the normal mouse tail without any injection. To elucidate further the site of origin, the spinal cord was dissected in the mid-thoracic region under light ether anesthesia; several hours later, amidone-HCl, morphine sulfate and strychnine sulfate in the previous doses were injected, using the same route of administration. Whereas amidone-HCl and morphine sulfate did not induce any tail reaction under these conditions, the injection of strychnine sulfate still elicited elevation of the tail, sometimes in connection with general tetanic convulsions; sometimes only the elevation of the tail took place. Further, after decerebration of mice, general decerebrate rigidity with convulsions and elevation of the tail were seen similar to that after amidone-HCl, morphine sulfate or strychnine injections. The elevation of the tail of white mice seems to be a normal reflex phenomenon, perhaps originated in the spinal cord. The development of this reaction seems to be facilitated either by increased activity along the cerebrospinal pathways (due to amidone or morphine) or by increased excitability of the spinal cord (due to strychnine).

The use of natural and synthetic fat emulsions for intravenous feeding. HARRY H. LEVEEN (introduced by FRANK CO TUI). *Department of Surgery, New York University College of Medicine.* A safe, metabolizable solution of fat for intravenous use would be significantly valuable in clinical nutrition. In preparing emulsions for intravenous use, extremely small particle sizes are essential to prevent fat embolization and to stabilize the emulsions. Since it is impossible to make emulsions of natural fats with satisfactorily small particle size by mechanical methods, emulsifying agents must be added to lower the surface tension between the fatty and the aqueous phases. However, fat films are an integral part of the cell membrane and since the emulsifying agents tend to make the fat film soluble, they disrupt cell and capillary membranes. Even non-toxic emulsifying agents, used heretofore, produce hemolysis of red cells and increased capillary permeability. The latter is especially noticeable as pulmonary edema. Death ensues from the sudden development of a shock-like state characterized by a rapidly falling blood pressure. For these reasons, it would seem impossible to prepare satisfactory emulsions of natural fats. Synthetic fats will form stable emulsions of extremely small particle size without emulsifying agents. The synthetic fats under investigation by the author include the esters of the higher fatty acids with sugars and polyhydric alcohols, the very short chain fatty acids and their esters, and the dicarboxylic fatty acids. The intravenous use of some of these synthetic fats is discussed.

The physiological disposition of procaine in man. PHILIP A. LIEF (by invitation), RAYMOND POET (by invitation) and BERNARD B. BRODIE. *Departments of Anesthesiology and Biochemistry, New York University College of Medicine, and New York University Research Service, Goldwater Memorial Hospital, N. Y., N. Y.* Procaine has recently been administered intravenously to relieve pain and to remedy spontaneous cardiac arrhythmias caused by cyclopropane. The fate of Procaine in man was studied for information concerning the mechanism of these actions.

Procaine hydrochloride was given intravenously to various subjects. Procaine, diethylaminoethanol and p-aminobenzoic acid were identified in the urine. These substances were then estimated in urine and plasma. Urinary excretion accounts for about one per cent of administered procaine. Therefore, the largest fraction of procaine undergoes metabolic alteration. P-aminobenzoic acid (and derivatives) excreted is equivalent to about 80 per cent of procaine. Diethylaminoethanol excreted is equivalent to about 25 per cent of

administered procaine. When diethylaminoethanol is administered intravenously only about 25 per cent is recovered in the urine. The nature of the metabolic alteration of diethylaminoethanol has not been established. Procaine disappears rapidly from plasma, only traces being found even during injection. Appreciable concentrations of diethylaminoethanol and p-aminobenzoic acid are found. These results indicate that procaine rapidly hydrolyzes *in vivo* to p-aminobenzoic acid and diethylaminoethanol. Procaine also hydrolyzes rapidly in human plasma *in vitro*. The rapidity with which procaine disappears from the body and the relative persistence of diethylaminoethanol suggest the possibility that the latter may be the active agent. Preliminary experiments show that diethylaminoethanol is much less toxic than Procaine and may be administered orally in relatively large doses. It is being tested for its analgesic properties and for its effect on the heart arrythmias of cyclopropane anesthesia.

Absorption of salicylates from intestinal loops in the dog. E. WILLIAM LIGON, JR. (by invitation), ROBERT A. MADDEN (by invitation) PAUL L. DAVIS (by invitation) and PAUL K. SMITH. *From the Department of Pharmacology, The George Washington University School of Medicine, Washington, D. C.* It has been reported that sodium bicarbonate increases the rate of absorption of concurrently ingested salicylates. In this study of salicylate absorption, to eliminate the hastening effect of alkali on gastric evacuation and more accurately control the pH of the gut contents, trained unanesthetized Thiry-Vella jejunal loop dogs were employed. Solutions containing 200 mgm. per cent salicylic acid were made isotonic by adding sodium chloride and/or sodium bicarbonate (pH 2.5 to 7.8) and passed through the gut loops at about 300 cc. per hour. Plasma and urine content of salicylate were determined spectrophotometrically at two hours. Fifty-one experiments on three dogs showed better correlation of plasma level with the pH of the outflowing perfusate than with the lower pH of the inflowing solution. Maximum plasma levels of approximately 10 mgm. per cent were obtained at outflow pH 5.0 to 5.8, with values decreasing to approximately 4 mgm. per cent at either extreme. Urine content of salicylate was an insignificant correction factor in effect of pH on rate of absorption. It has been shown that acetylsalicylic acid injected intravenously in the dog is rapidly hydrolyzed. Experiments were designed to demonstrate whether or not acetylsalicylic acid is hydrolyzed prior to absorption, using aspirin solutions in the same dogs in a similar manner. No greater concentration of free salicylate was found in this perfusate (pH 5.4) than in control solutions, suggesting that aspirin is hydrolyzed after absorption.

The toxicity of some surface active agents on the frog heart. ALFRED G. LISI, (introduced by CHARLES M. GRUBER). *Department of Pharmacology, Jefferson Medical College, Philadelphia, Pa.* In an effort to determine the possible effect of residues left on glassware cleaned with the aid of the newer surface active agents, a study was made of the action of certain commercial preparations on the excised, perfused frog heart. An all-glass cannula was used which has a side arm for maintaining a constant level of solution. The following preparations were employed: sodium lauryl sulfate (Du Pont's "sodium 'lorol' sulfate PT"), a polymerized ethylene oxide condensation product (Igepal CA Extra, an aryl alkyl sulfonate (Nacconol NRSF) and a sorbitan monooleate polyoxyalkalene derivative (Tween 80). The most useful criterion was found to be the observation of a minimal decrease in the height of contraction as recorded on a kymograph. Ringer's solution was perfused through the heart during control periods and between applications of the surface active agent. Appropriately diluted solutions of the test substances in Ringer's solution were applied to the heart for six minute periods. The inhibition regarded as "minimal" was such that it could be readily seen by comparison with the preceding and following control periods. Concentrations of solution could only be recorded in terms of per cent since the materials tested are not pure compounds. In general terms, the surface active agents tested were toxic in the range of 0.0005% – 0.005% solutions. For comparison, potassium dichromate was tried and its comparable toxicity was around 0.01% – 0.02%.

A simplified method of evaluating dose-effect experiments. J. T. LITCHFIELD, JR. and F. WILCOXON (by invitation). *Chemotherapy Division, Stamford Research Laboratories, American Cyanamid Company, Stamford, Connecticut.* By means of tables and nomographs to be presented, an approximate solution of dose-effect curves can be made quickly and easily by the use of a slide rule and logarithmic probability paper. No elaborate computation and no use of logarithms or probits is required. The method includes a test of the goodness of fit of the line to the data by means of a $(Chi)_2$ test, as well as the estimate of the median effective dose ($E.D._{50}$) and its 19/20 fiducial limits. The method makes possible also the estimation of the 19/20 fiducial limits of S. Since S with its limits is a function of the slope constant (b) and its standard error, the degree of parallelism between two assays can be tested. If the curves of two assays are parallel within the limits of error, the ratio of potency may be computed and the 19/20 fiducial limits established.

Anticonvulsant actions of trimethadione-phenobarbital and trimethadione-diphenylhydantoin

combinations. S. LOEWE. *Department of Pharmacology, University of Utah School of Medicine, Salt Lake City, Utah.* (a) Protection against metrazol convulsions and (b) abolition of tonic hindleg extension phase of maximal electroshock seizures were measured in various species, by determining the protective doses (PD_{50}) of the individual drugs and the isoboles, as previously described, of the drug combinations. The anticonvulsants trimethadione (T), phenobarbital (Ph) and diphenylhydantoin (D) were injected intraperitoneally, metrazol was given subcutaneously (rats, mice) or intravenously (cats). (a) Whereas no *antimetrazol* synergism of T+Ph was demonstrable in mice and cats or of T+D in rats, T moderately synergized Ph in rats and D in mice, but only when the dose ratios T/Ph and T/D were low. (b) *Antishock* synergism of marked degree was demonstrable in all cases studied (T+Ph in rats and cats; T+D in rats). However, the synergism of T+Ph in rats was limited to high T/Ph dose ratios, whereas antagonism prevailed in the range of small T/Ph ratios. T and D (single D dose, four hour interval) synergized mutually in all dose ratios; also repeated D doses were markedly synergized by as little as $\frac{1}{3}$ PD_{50} of T. Since efficacies in petit and grand mal are correlated with antimetrazol and antishock activity, respectively, these results may be instructive for selecting optimal therapeutic combinations. They suggest that in petit mal a T regimen might not profit from small Ph or D additions, but vice versa; in grand mal, a T regimen might be improved by small Ph additions—but not vice versa—, and T+D combinations in all ratios might offer advantages.

The response and the mechanism of the response of the duodenum to morphine. TED A. LOOMIS (introduced by JAMES M. DILLE). *Department of Pharmacology, University of Washington School of Medicine, Seattle, Washington.* The muscular response of the duodenum to intravenously administered morphine was studied in intact anesthetized dogs. Apparatus designed to measure simultaneously the changes in circular and longitudinal muscle activity was used. The predominant effect of morphine on the circular muscle group was found to be an increase in its many phases of activity. The predominant although not completely consistent effect on the longitudinal muscle group was found to be a decrease in its many phases of activity. The type of response was found to be independent of the original state of tonus and of the dose over a range of 0.1 to 1.0 mgm. per kg. In the presence of a barium spasm, morphine produced little or no alteration of muscle activity. In the presence of physostigmine or atropine the response to morphine was similar to the normal response produced by morphine. The data obtained indicates that the site of action of morphine is distal to the acetylcholine-choline esterase mechanism resulting in stimulation of the circular muscle group and depression of the longitudinal muscle group.

Comparative effects of sparteine and quinidine on isolated frog heart. GO LU (Fellow, American Bureau for Medical Aid to China Inc.; by invitation). *From the Department of Pharmacology and Therapeutics, Stanford University School of Medicine, San Francisco 15, Calif.* Using a modified assembly of the Hartung-Clark double cannula and the Straub ventricle methods sparteine and quinidine were compared on known characteristics of the isolated frog heart. The predominant effects of sparteine were purely depressant, qualitatively resembling quinidine in all essential respects, i.e., bradycardia, typical lengthening of the refractory period (also demonstrated on isolated skeletal muscle), relaxation of cardiac muscle, decreased contractility and cardiac output, abolition of extrasystoles, and prevention and arrest of auricular fibrillation. These changes were either abolished or prevented by atropinization, and therefore were muscular in origin. The apparent initial stimulation of sparteine indicated by moderate increases in amplitude of contractions and stroke and minute output (not produced by quinidine) is presumably due to the slowing in rate and relaxation of cardiac muscle. Ready conversion of this initial action to a marked depression, tendency to diastole, and lack of benefit of sparteine on failing hearts were further evidences supporting the slowing mechanism of "stimulation," simulated in all essential respects by cooling of the sinus node. The sparteine arrested heart was readily reversed by washing with Ringer's solution or addition of epinephrine, calcium, and caffeine, but not so readily quinidine arrested hearts. Toxic concentrations of sparteine were somewhat higher than those of quinidine. According to these results, sparteine satisfies the essential requirements of a quinidine-like drug, with a somewhat lower toxicity, a freedom from cinchonism, and a higher solubility permitting possibly better absorption and easier adjustment of dosage. Sparteine seems worthy of further clinical trial.

Some pharmacological actions of isomers of methadon. F. P. LUDUENA, LLOYD C. MILLER, ESTELLE ANANENKO (by invitation) and J. D. FRICK (by invitation). *From Biology Division, Sterling-Winthrop Research Institute, Rensselaer, N. Y.* Using the radiant heat method of Ercoli and Lewis the analgesic potencies of *l*- and *d*-methadon, (6-dimethylamino-4,4-diphenyl-3-heptanone), and the *dl* form of a structural isomer, ("Win 1783", 6-dimethylamino-4,4-diphenyl-5-methyl-3-hexanone), were determined by subcutaneous injection in rats. The approximate threshold analgesic doses were 1, 20 to 30, and 4

mg/kg., respectively. Thus the *l*-isomer is responsible for most of the analgesic action of racemic methadon. This observation confirms the report of Thorp, Walton and Ofner on *l*- and *dl*-methadon.

Win 1783, which differs from methadon only in the position of the methyl group, is about half as active as *dl*-methadon, but the respective therapeutic indices are about the same, as judged from toxicity data described elsewhere in these Proceedings (Hoppe and Miller). On barbitalized dogs quantitative and qualitative differences were observed between the actions of *d*- and *l*-methadon given intravenously. The first injection of *l*-methadon lowered the blood pressure; subsequent injections produced a moderate rise after a fleeting fall. The *d*-isomer produced only a depressor effect. Other pharmacodynamic effects of these analgesics on anesthetized and unanesthetized dogs will be discussed.

The protective action of BAL on experimental poisoning by lead, tungsten, copper and Paris green. LEHMAN M. LUSKY (by invitation), HERBERT A. BRAUN and EDWIN P. LAUG. *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington 25, D. C.* We previously reported that BAL enhanced the toxic effect of lead salts administered by intramuscular or intraperitoneal injections to rabbits. This led to investigations to determine whether the increased toxicity was due to the formation of a toxic complex or systemic mobilization. In the present studies single intravenous doses of lead acetate (20–40 mg./kg.) were administered intravenously to young rabbits. Initial treatment of BAL (30 mg./kg.) was followed by descending subcutaneous doses during the 30-day experiment. Comparison of the mortality curves of control and BAL-treated animals indicates that BAL reduced the toxicity of intravenously administered lead acetate by 50%. The bone of BAL-treated animals contained smaller concentrations of lead than the controls. Control animals given 110 mg./kg. sodium tungstate intramuscularly died within 5 hours. BAL (30 mg./kg.) intramuscularly in the opposite leg within 20 minutes after the tungstate injection saved all rabbits from this dose of sodium tungstate. The effect of BAL on experimental copper poisoning is striking. Fifteen control rabbits intravenously injected with cupric sulphate (20 mg./kg.) died within 30 minutes. BAL (30 mg./kg.) intravenously after the injection of cupric sulphate saved 14 of 15 rabbits. The antidotal effect of BAL against Paris Green was demonstrated in rabbits. Orally, 25 mg./kg. was fatal to 15 of 18 animals, while 13 of 18 were saved with BAL started at 30 mg./kg. twice a day and continued in descending doses for 8 days.

Influence of benadryl and pyribenzamine on the neuromuscular system of rats. DAVID I. MACHT and THOMAS HOFFMASTER (by invitation). *Division of Pharmacology, Laboratories of Sinai Hospital, Baltimore.* Inasmuch as these two drugs produce occasional reactions particularly on the part of the Nervous System, studies were made on the behavior of white rats by methods described elsewhere (Macht D. I. Experimental Medicine and Surgery, 1943, Volume I, Page 260). Three sorts of experiments were performed: (1) on the behavior of rats in a circular maze giving data on neuromuscular activity and on discrimination or "choice" reactions: (2) on the coordination of rats running across a tightly stretched horizontal rope: (3) on neuromuscular work performed by rats climbing a vertical rope. Doses of both Benadryl and Pyribenzamine ranged from 0.1 mg. to 2 mgs. Injections were made either intraperitoneally or subcutaneously. As little as 0.2 mgs. of either drug produces a depression in activity within 15 minutes. Doses of 0.5 mg. and more cause marked depression. When larger doses (0.5–2.0 mgs.) are injected in rats weighing 150–200 gm., the maze performance is impaired more than the rope walking and climbing, thus pointing to the C.N.S. being more affected than the muscular responses. Even after doses of 5 mgs. the animals recovered in a few days.

Phytotoxic effects of normal, pathological, and irradiated blood sera. DAVID I. MACHT and MARCUS OSTRO (by invitation). *Departments of Pharmacology and Radiology, Sinai Hospital, Baltimore.* This being the 25th anniversary of the senior author's researches in Phytopharmacology, his experiences with thousands of blood specimens are reviewed and analyzed, and new discoveries are here presented. Blood sera of all higher animals, except reptiles, give normal indices of root-growth (ranging 70% to 75%) for Lupinus albus seedlings. Reptilian sera are toxic. Sera of Virus diseases are weaker than normal sera. The following conditions yield specific toxic indices, which are useful in diagnosis. Menstruation, Pernicious Anemia, Pemphigus, Leprosy and Trachoma. P.A. serum is phytotoxic; all other anemias and leukemias are not. It is detoxified in quartz by ultraviolet rays of a mercury vapor lamp. So are patients in vivo. Pemphigus serum is unaffected by ultraviolet, but detoxified by specially filtered very short x-rays. Such deep x-rays over the liver and spleen produce favorable therapeutic effects and have saved several moribund cases. (Urological and Cutaneous Review November, 1947.) Menotoxin, Trachoma and Leprosy sera are unaffected by roentgen rays. Numerous serum specimens from psychotic patients revealed that all psychoses whether organic or functional are phytotoxic; and are detoxified *in vitro* by special x-rays. Animals receiving penicillin affect root-growth, and patients' blood levels can be estimated phyto-

pharmacologically. "Vernalized" or "Iarovized" seedlings yield a specific reaction with Psoriasis blood not shown by normal plants (Edwin C. Miller, Plant Physiology, Second Edition p. 1091).

The distribution, excretion, and blood levels of antimony following administration of antimonials to various mammalian species. THOMAS H. MAREN and GILBERT F. OTTO (introduced by E. K. MARSHALL, JR.). The trivalent and pentavalent antimonials in current clinical and experimental use show wide differences in their distribution and excretion; there are also significant differences in the behavior of these drugs in various species of animals. Compounds used included among others tartar emetic, monosodium antimony thioglycollate, "fuadin", and "solustibosan". Pharmacological data was obtained for the white mouse, hamster, cotton rat, guinea pig, rabbit, dog, and man. In these species natural or experimental infection of several types have long been treated with antimonials; in the present study uninfected animals as well as those with filariasis and schistosomiasis have been used in an attempt to provide a pharmacological basis for therapy. Following intraperitoneal or intravenous injection the trivalent compounds give rise to deposition of antimony principally in the thyroid gland and the liver. Pentavalent compounds result in antimony concentration in the spleen, usually with lesser amounts in the liver. Quantitative differences were observed with different species. In dog and man, following trivalent antimony, there is approximately 5 times as much metal in red cells as in plasma. After pentavalent compounds, on the other hand, plasma concentrations at the peak were 10 times that of the cells. Excretion was extremely variable with the species. In hamsters, mice, and white rats receiving trivalent antimony, the metal was eliminated mainly in feces. In guinea pigs, rabbits, dogs, and in man it is excreted largely in urine. In all species receiving the pentavalent drugs, the metal was rapidly eliminated in the urine.

Antagonism of nicotine and atropine. AMEDEO S. MARRAZZI. *Department of Pharmacology and Therapeutics, Wayne University College of Medicine, Detroit, Michigan.* Marrazzi (J. Pharmacol. **65**:18; 1939, Fed Proc. **6**:354; 1947) has shown that atropine is capable of blocking transmission in sympathetic ganglia. Therefore, a distinction, such as a so-called nicotinic action as opposed to a muscarinic, based on experimental procedures dependent on an alleged immunity of ganglia and nicotine to atropine requires re-examination. The rise in blood pressure caused by nicotine is considered a characteristic action. It seemed desirable, therefore, to determine whether it may be modified by the prior injection of atropine. Such an effect on the circulatory system, which is controlled by so many quite variable factors including compensatory reflexes is quite apt to be masked. Accordingly, we found a constant pressor effect of nicotine and its depression by the prior intravenous injection of atropine in only the occasional experiment where conditions appeared to have fortuitously favored its detection. However, on eliminating the vagi and the carotid sinuses more regular results are obtained showing a control rise on the intravenous injection of nicotine, the absence or reduction in the expected rise due to nicotine when it is injected after atropine and a subsequent control rise again with the nicotine alone. The masking of the nicotine-atropine antagonism on circulation and on other effector systems, the mechanism of the fall in blood pressure after intravenous atropine and the bearing of the nicotine-atropine antagonism on concepts of cholinergic transmission will be discussed.

Synthetic curare compounds. III. d-N-methyl-chondrodendrine iodide and d-N-methyl-O-methyl-chondrodendrine iodide. DAVID FIELDING MARSH, CLARK K. SLEETH (by invitation), and ELDON B. TUCKER (by invitation). *Departments of Pharmacology and Medicine, West Virginia University School of Medicine, Morgantown, W. Va.* d-Chondrodendrine was isolated from Pareira brava. The curariform activity of its derivatives, d-N-methyl-condrodendrine iodide ("bebeerine dimethiodide", CM), and d-N-methyl-O-methyl-chondrodendrine iodide ("beberine methylether methiodide," CMMeO), was determined in comparison with the corresponding isomers, d-tubocurarine chloride pentahydrate (dT), and d-O-methyltubocurarine iodide trihydrate (dTMeO). The relatively high curariform activity, lack of undesirable side effects, and ready availability of these chondrodendrine derivatives make their clinical investigation seem reasonable.

	dT·Cl	dTMeO·I	CM·I	CMMeO·I
Albino rats				
LD 50	0.27 (0.22)*	0.032 (0.022)	0.55 (0.39)	0.37 (0.27)
Rabbits				
Head drop 50	0.12 (0.10)	0.016 (0.011)	0.23 (0.16)	0.11 (0.08)
Holaday head drop	0.15 (0.12)	0.020 (0.014)	0.27 (0.19)	0.13 (0.09)
LD 50	0.35 (0.28)	0.040 (0.027)	0.40 (0.28)	0.28 (0.17)
Cat gastrocnemius muscle				
Equivalent paralysis	0.05 (0.04)	0.007 (0.005)	0.40 (0.28)	0.05 (0.04)
Man				
Head drop	0.15 (0.12)	0.030 (0.020)	0.60 (0.43)	0.15 (0.11)

* All doses given in milligrams/kilogram body weight. Dose of contained curariform ion given in parenthesis.

Preliminary data on rat feeding with beryllium. ELLIOTT A. MAYNARD (by invitation), WILLIAM L. DOWNS (by invitation), and HAROLD C. HODGE (by invitation) (introduced by HARVEY B. HAAG. At a dietary level of 5%, Be sulfate and Be carbonate completely inhibited growth of rats and caused some mortality. A rickets-like condition occurred in these rats. Food intake was reduced by about 50%, but this did not account for all growth inhibition as shown by a paired feeding experiment. Return to stock diet after a prolonged period of ingestion of Be salt allowed rapid but not complete growth recovery; a healing of the rickets occurred in a few days. In turn this was followed by the appearance of a radio-translucent area extending into the shaft of the bone from the metaphyseal space. Be sulfate was more toxic to old rats than to weanlings, but with Be carbonate mature rats seemed to be more resistant than weanlings. Be carbonate at a level of 2.3% of the diet caused some weight depression in rats. Be metal at a 10% dietary level and Be oxide at a 5% dietary level did not affect the growth of the rats.

Some aspects of the action of curare on the central nervous system. E. L. MCCAWLEY. *From the Laboratories of Pharmacology and Toxicology and the Laboratory of Neuro anatomy, Yale University School of Medicine, New Haven, Connecticut.* Cats were used in the majority of experiments reported here; although dogs and rabbits were also employed. Records were made of the cortical electroencephalogram, cerebrospinal fluid pressure, the electrocardiogram and blood pressure. In all cases artificial respiration, 23 strokes per minute, was supplied through a tracheal cannula or endotracheal catheter. Curare, "Intocostrin" and d-tubocurarine chloride, and tetraethyl ammonium bromide were used. Since the effect of these drugs is transitory a constant rate of injection was employed such that the rate of administration exceeded the rate of elimination or destruction. By varying this rate of injection it was found that the appearance of symptoms and events followed a parabolic function of time and total dosage. In the unanesthetized animal doses of curare less than those required to produce paralysis of the intercostal muscles produced clonic convulsions. The EEG was characterized by high frequency spikes of considerable voltage, i.e., similar to that of grand mal. Tetraethyl ammonium elicited the same effect, as did also very small doses of curare placed in the cisterna magna. This phenomenon was prevented by the depressant drugs pentobarbital and thiopental and also by morphine. In the anesthetized animal cortical depression from curare appeared at 200–300 units/kg. body weight and the rate of administration exceeded 0.5 cc./minute. Cortical brain waves were depressed for brief periods of time and finally disappeared entirely (isoelectricity). However, although the cortical brain waves were eliminated, trains of spikes 14–18/second, characteristic of thallamic activity (or neighboring structures) appeared. Animals whose cortical activity was depressed by curare could not be revived. Although in certain animals the heart was kept beating by artificial respiration for 20 hours and the drug eliminated, there was no return of brain function. Physostigmine, neostigmine, guanidine, peripheral anticurare agents, had no demonstrable central effect. "Metrazol" alone produced a temporary return of cortical activity.

The action of p-aminosalicylic acid (PAS) in experimental tuberculosis. W. T. MCCLOSKY, M. I. SMITH and J. E. G. FRIAS (by invitation). *Division of Physiology, National Institute of Health, Bethesda, Maryland.* The acute and chronic toxicity and fate of p-aminosalicylic acid (PAS) were studied in rats, rabbits and guinea pigs and its effectiveness in experimental rabbit and guinea pig tuberculosis ascertained. The acute toxicity of PAS is low. Rats tolerated 2.0 gm/kg. when injected intravenously, and in guinea pigs on oral administration the approximate LD_{50} was 3.0 gm/kg. Continued oral administration to guinea pigs in daily doses of 0.5 gm/kg. showed cumulative action since 50% of the animals died after 18 doses. The compound is well absorbed from the gastro intestinal tract, is retained for 3 to 6 hours, and is nearly completely excreted in the urine within 24 hours, for the most part in the conjugated form, probably as the acetylated product. In rabbits infected with .01 mg. bovine strain (Ravenel) tubercle bacilli i.v. and treated with PAS in doses of 0.2 to 0.5 gm/kg. daily the average extent of tuberculous involvement for a group of six animals was 3.0 as compared with 5.1 for the untreated controls, whilst a similar group treated with streptomycin, 40 mg/kg/day, showed no lesions in five of the animals and minimal lesions in one. A group of animals treated with both drugs had no evidence of tuberculosis. Four groups of guinea pigs, ten each, inoculated intraperitoneally with 0.5 mg. tubercle bacilli human strain A27 and treated with PAS 0.3 to 0.5 gm/kg/day alone or in combination with 20.0 mg/kg/day of streptomycin showed an average extent of tuberculous involvement as follows: Untreated controls 10.7, treated with PAS 7.4, treated with streptomycin 1.5, treated with both drugs 1.3. Thus the chemotherapeutic effectiveness (ratio of T.B. in controls/treated) of PAS was 1.4, of streptomycin 7.1 and of combined therapy 8.2, which is no greater than the sum of effects from the individual components. It is concluded that the chemotherapeutic efficacy of PAS in tuberculosis is slight compared with the sulfones, and unlike the sulfones it gives no indication of potentiation with streptomycin.

Acute vapor toxicity to mice and local irritant effects in the rabbit of methyl pentadiene. W. A. McOmie (introduced by Hamilton H. Anderson). *Division of Pharmacology and Experimental Therapeutics, University of California Medical School and College of Pharmacy, San Francisco, California.* The material used was a mixture of approximately 85% of 2-methyl-1,3 pentadiene and 15% of 4-methyl-1,3 pentadiene. Stock white mice were exposed in a chamber to the vapors of this low boiling (75°C.) olefinic hydrocarbon by diluting nitrogen and vapor mixture with different rates of flow of air, so as to obtain a dosage mortality relationship (see table). The periods of exposure varied from 30–270 minutes and the concentration of vapor from 3–150 mgm./l.

Dosage-mortality relationship upon exposure of mice to vapors of methyl pentadiene*

ct product (mgm./l × minutes)	No. of mice dying within 96 hours/Total no. mice used
3780	0/6
8400	2/6
12000	8/12
18400	4/6

* $L(ct)_{50}$ = Product of concentration × exposure period causing 50% mortality.

An $L(ct)_{50}$ of 10,000 mgm.min./l was estimated from these values using the graphic method of Miller and Tainter (Proc. Soc. Exp. Biol. and Med. 57: 261, 1944). If one-half the $L(ct)_{50}$ can be assumed as a safe concentration, 10 mgm./l. (3000 p.p.m.) should be the threshold concentration for an 8-hour exposure in mice. Effects in mice upon vapor inhalation resembled that of other anesthetic agents, e.g., hyperexcitability and ataxia preceding anesthesia. If the exposure was sufficiently prolonged, death ensued due to respiratory failure. The only involvement seen in mice upon autopsy was lung congestion. These same general effects and a similar order of toxicity have been reported for 1,3 butadiene by Carpenter et al. (Jour. Ind. Hyg. and Toxicol. 25: 255, 1944). Local effects upon application of the undiluted material to the shaved, non-abraded skin of the rabbit consisted of immediate vasodilation, with a subsequent erythema which persisted for 24 hours; drying of the skin and superficial sloughing occurred after 5–7 days. The undiluted material applied to the eye of the rabbit caused a conjunctivitis that persisted for 24 hours but was not followed by permanent damage.

The anti-fibrillation action of papaverine and its value in cardiac resuscitation after chloroform-adrenaline ventricular fibrillation. K. I. Melville. *Dept. of Pharmacology, McGill University, Montreal, Canada.* Dogs anesthetized with sodium pentobarbital were used. Artificial respiration was maintained throughout all experiments by means of a Starling pump. Blood pressure and electrocardiograms (Lead 11) were recorded. In normal and vagotomized animals, the intravenous injection of 5 to 10 mgm. per kgm. of papaverine hydrochloride during chloroform administration, can protect the heart and prevent the development of ventricular fibrillation following intravenous injection of 0.02 mgm. per kgm. of adrenaline. This protective effect wears off in 20 to 30 minutes, but can be repeated. After induction of chloroform-adrenaline ventricular fibrillation in 12 experiments, the intracardiac injection of papaverine (5 mgm. per kgm.) in conjunction with cardiac massage, arrested the fibrillation and restored a normal coordinated ventricular beat. At this stage, the heart is dilated, the beat is feeble, and the ventricular rate considerably slowed. Injection of adrenaline (intracardiac) with brief massage of the heart, can then accelerate the heart rate and restore the circulation. This procedure might be of some aid in cardiac resuscitation after ventricular fibrillation. It is suggested that the anti-fibrillation action of papaverine is due primarily to its coronary dilator action. Finally, repeated injections of papaverine during chloroform inhalation can induce circulatory failure associated with ventricular extrasystoles and fibrillation. This condition can however be restored to a normal coordinated beat by cardiac massage.

The effect of some glycerin esters upon the blood pressure, uterus, and respiration. Rafael Méndez and Ernesto Sodi (by invitation). *From the Department of Physiology and Pharmacology of the National Institute of Cardiology, Mexico City.* The effects were studied of the acetyl esters of glycerin, monoacetin, diacetin and triacetin and the sulfoesters trisulfoglycerin ($C_3H_8O_{12}S_3$) and tetrasulfodiglycerin ($C_3H_7O_8S_2—O—C_3H_7O_8S_2$) upon the blood pressure, uterus "in situ" and respiration. Eviscerated, adrenalectomized cats under Dial anesthesia were used. Monoacetin up to 0.25 cc./kg. caused a small and brief decrease (12–20 mm. Hg) in blood pressure with an uncertain effect upon the uterus and an increase in respiration. The same doses of diacetin caused a profound decrease in blood pressure (80–100 mm. Hg) lasting 3–5 minutes, a relaxation of the uterus lasting 10–15 minutes and an increase in the rate and amplitude of respiration. Triacetin, 0.05–0.1 cc./kg. caused the same effects as Diacetin (0.25 cc./kg.). Thus the blood pressure decreasing effect increases with acetylation of the hydroxyl groups of glycerin. Trisulfoglycerin and tetrasulfodiglycerin in doses of 15 mg./kg. caused a pronounced increase in blood pressure lasting for 10–15 minutes, and a decrease in the rate and amplitude of respiration. These experiments were conceived to find in the glyceryl radical an effect which would explain the high

potency of nitroglycerin as compared with the heavier organic "nitrates" and with sodium nitrite. The results with the acetines support this assumption. Work is continued with other esters. The results with the sulfo-esters do not invalidate the idea, as the introduction of the SO_3 usually cancels or modifies the effect of the original compound. (Rojahn and Giral: Productos Químicos Farmacéuticos, Editorial Atlanta, México, 1942).

Comparison of digitalis glycosides with their genins in man. WALTER MODELL, NATHANIEL T. KWIT (by invitation), CONRADO DAYRIT (by invitation), S. J. SHANE (by invitation), and HARRY GOLD. *Department of Pharmacology of Cornell University Medical College, Cardiovascular Research Unit of the Beth Israel Hospital, and Cardiac Service of the Hospital for Joint Diseases, New York.* There are some reports in animals showing that the aglycones of the digitalis series possess the typical action of the glycoside on the heart, but that with the removal of the sugar fraction from the glycoside, the potency is usually reduced, the persistence of action is markedly shortened, and a convulsant action may become prominent. How these matters stand in the human subject was tested in patients with congestive failure and auricular fibrillation, using the changes in apex rate as the chief guide. The method was similar to that described in several of our previous studies comparing digitalis materials in man. The final analysis depends on the results obtained in 6 patients in each of whom genins were compared with corresponding glycosides by intravenous injection, digitoxigenin with digitoxin, digoxigenin with digoxin and lanatoside C. The single doses varied from 0.75 to 2 mg. The genins behave like their glycosides in slowing the ventricular rate in patients with auricular fibrillation. As has been shown in animals, in man also their action on the heart is conspicuously weaker when equal doses by weight are used, the difference being markedly increased when the compounds were compared on a molecular basis. There are 2 other differences; the genins develop their full action within a few minutes as against several hours for the corresponding glycosides; the effect of the genins disappear within 30 minutes to a few hours while that of the corresponding glycosides lasts from many hours to many days.

Pharmacologic studies of the neurotoxic properties of streptomycin. HANS MOLITOR and SAMUEL KUNA (by invitation). *Merck Institute for Therapeutic Research, Rahway, N. J.* Even pure streptomycin produces certain toxic manifestations among which a selective effect upon the vestibular system is outstanding. The latter may be regarded, therefore, as an intrinsic property of the streptomycin molecule. Clinical reports indicate, however, that patients receiving equal doses of different samples of streptomycin may show variations in neurotoxic reactions greater than explainable by differences in individual patient sensitivity. We have previously shown that streptomycin lots causing a high incidence of clinical neurotoxicity also were highly neurotoxic when injected intracisternally into rabbits. However, since the reaction following intracisternal injection of a drug is not necessarily the same as that produced by repeated intramuscular injections (the usual mode of streptomycin administration in man) we compared in rabbits and cats the effect of single intracisternal injections with that of repeated subcutaneous injections. Twenty streptomycin samples ranging in potency from 85 units/mg. to 800 units/mg. were examined in some 200 cats and 500 rabbits. The doses employed were from 750–4000 units/kg. intracisternally and from 25,000–900,000 units/kg./day intramuscularly. Samples which were highly neurotoxic upon single intracisternal injection also possessed high neurotoxicity when given repeatedly subcutaneously. Thus, a sample of streptomycin hydrochloride producing neurotoxic signs upon intracisternal injection of 500 units/kg. produced similar signs within 20 days upon daily subcutaneous injection of 50,000 units/kg. while a sample with an acute neurotoxicity of 2000 units/kg. required from 60 days upward for a comparable effect.

Rate of disappearance from the blood and distribution in tissues of 3-(orthotoloxyl)-1,2-propanediol (myanesin). JAMES L. MORRISON, ARTHUR P. RICHARDSON and HARRY A. WALKER (by invitation). *Department of Pharmacology Emory University, Georgia.* Using the diazotization method of Titus, Ulick, and Richardson (Journ. Pharmacol. & Exper. Ther. (in press) the plasma levels of 3-(orthotoloxyl)-1,2-propanediol (myanesin) were determined in a series of dogs infused with varying concentrations of the agent. Blood levels were followed until disappearance from the blood of an intravenous priming dose of 60 mgm./kgm. In most cases the same dogs were used again, after suitable intervals, for intravenous infusion using a priming dose of 60 mgm./kgm. followed by the infusion with myanesin at the rate of 1 mgm./kgm./min. for two hours. Subsequently this was repeated using a rate of infusion of 2 mgm./kgm./min. Blood levels were determined at suitable intervals during infusion and until the drug disappeared from the blood stream on cessation of the infusion. Infusion was through a cannula in the femoral vein. Blood samples were taken from the jugular vein. Following the last infusion certain animals were sacrificed by injection of air and organs were removed and myanesin content was determined by the above method. Concentrations of myanesin appear to be much higher in brain, pancreas, kidney, spleen, in a descending order and

only slightly higher than plasma in muscle, lung and liver. The concentration of myanesin in spinal fluid and saliva was slightly lower than that in plasma. Blood level curves obtained by this method indicate that doses as high as 60 mgm./kgm. can be disposed of by the body in a matter of 30–60 minutes. By infusing with myanesin after such a dose of 60 mgm./kgm. an infusion rate can be estimated which will maintain a constant level in the blood. Since infusion rates of 1 mgm./kgm./min. will not maintain a constant level and rates of 2 mgm./kgm./min. leads to accumulation of myanesin the maintenance level must lie somewhere between.

Studies with the polyoxyethylene derivatives of sorbitan partial esters (Tweens). RUTH MUSSER (introduced by JOHN C. KRANTZ, JR.). *Department of Pharmacology, Univ. of Maryland, School of Medicine, Baltimore.* Tween 20, 40, 60 and 80 were studied in relation to their capacity to hemolyze red cells. Although the Tweens as a class were found to be hemolytic, their products of hydrolysis were nonhemolytic. Pancreatic lipase caused the hydrolysis of Tweens *in vitro*. The rate of the polyoxyethylene derivative of sorbitan was studied in humans with special reference to excretion and oxaluria.

Effect of procaine on cyclopropane-epinephrine cardiac arrhythmias. MARK NICKERSON. *Department of Pharmacology, University of Utah School of Medicine, Salt Lake City, Utah.* After pentothal induction, dogs were equilibrated to a 30 per cent cyclopropane-70 per cent O_2 anesthetic mixture for 30 minutes and then tested with an intravenous dose of 10 μgm./kgm. epinephrine. Procaine HCl (10 mgm./kgm.) administered 10 minutes prior to testing provides only slight protection against the epinephrine-induced cardiac irregularities. However, the same dose of procaine injected rapidly after irregularities develop will stop them as soon as the drug reaches the myocardium. Reversion to a sinus rhythm is accompanied by a prolongation, a reduction in amplitude and other changes in the QRS complexes of the EKG indicating altered myocardial conduction. Intravenous administration of 10 mgm./kgm. procaine HCl over a period of one minute considerably reduces the maximum frequency of A-V impulse transmission (reciprocal of the physiologically effective refractory period). Maximum reduction occurs near the end of the injection while a high concentration of procaine is being carried directly to the heart. Less than 40 per cent of the effect remains 3 minutes after the injection and it is almost completely gone within 10 minutes. The above results indicate that high concentrations of procaine are necessary to suppress epinephrine induced ectopic rhythms. They provide an explanation for the reported efficacy of procaine in arresting cardiac arrhythmias during anesthesia, but point to a very limited prophylactic value of this agent.

Dibenamine protection against cyclopropane-epinephrine cardiac arrhythmias. MARK NICKERSON and GEORGE M. NOMAGUCHI (by invitation). *Department of Pharmacology, University of Utah School of Medicine, Salt Lake City, Utah.* Three etiological factors proposed to explain cardiac irregularities produced by epinephrine in the presence of anesthetics are: (1) a direct effect of sud denly increased arterial pressure upon sensitized ventricles, (2) an inhibitory vagal action (secondary to increased arterial pressure) on the S-A node during the period of direct myocardial stimulation by epinephrine, and (3) a direct effect of epinephrine on the sensitized myocardium. All of these factors could be altered by Dibenamine. The nearly complete protection afforded by Dibenamine against cyclopropane-epinephrine arrhythmias is not diminished by constriction of the thoracic aorta in such a way as to reproduce the pressor effect characteristic of epinephrine in the absence of Dibenamine. The protection is thus unrelated to blocking of the epinephrine-induced rise in arterial pressure. Although strong electrical stimulation of the vagus produces a ventricular rhythm in the presence of both epinephrine and Dibenamine, the vagus is probably not an important factor in cyclopropane-epinephrine arrhythmias because bilateral vagotomy is not protective and reflex activation of the vagus does not alter Dibenamine protection. Dibenamine, therefore, appears to protect by acting directly on cardiac muscle, but its exact mode of action is still obscure. It does not have any direct effect on the heart detectable in the EKG, and does not alter the cardioaccelerator or inotropic effects of epinephrine. In addition, its "quinidine-like" effect on impulse conduction in cardiac muscle is too short in duration to account for the protection, and is also exerted by congeners which do not protect against cyclopropane-epinephrine arrhythmias.

Some applications of sequential analysis. R. H. NOEL and M. A. BRUMBAUGH (introduced by LLOYD C. MILLER). *Bristol Laboratories, Inc., Syracuse, New York.* Sequential analysis of laboratory results offers two major advantages to the technician, (a) defined risks of erroneous decisions, (b) reduction of the number of experiments required to arrive at decisions. Three illustrations are presented in this paper: (1) The amount of testing required to determine whether an experimental drug is superior to an established preparation. (2) The amount of testing required to determine whether a new antibiotic strain is superior to an established one. (3) The amount of testing required to assess the relative value of two assay methods.

The renal elimination of caronamide (4'-car-

boxyphenyl-methanesulfonanilide. HAROLD M. PECK, HORACE F. RUSSO, ELIZABETH K. TILLSON, WILLIAM S. WALLER (all by invitation) and KARL H. BEYER. *From the Department of Pharmacology, The Medical Research Division, Sharp and Dohme, Inc., Glenolden, Pa.* Caronamide (4'-carboxyphenylmethanesulfonanilide) is excreted as such and in an "altered", probably conjugated, form. The analytical methods of Sprague and Ziegler (J. Lab. Clin. Med., In press) and of Brodie, Levy and Bernstein (J. Pharmacol. **91:** 246, 1947) have been used in these experiments. Although these methods are reasonably accurate, the fidelity with which they measure "unaltered" caronamide (method of Brodie, et. al.) or total caronamide (method of Sprague and Ziegler) in urine or plasma of an animal to which the drug is administered does not appear to be certain. This increases considerably the desirability of several approaches to the problem of renal elimination of caronamide and the necessity for caution in interpretating the results. The results of the following types of experiments relating to the renal extraction, clearance, and urinary recovery of caronamide will be presented: 1) A comparison of the falling plasma concentration and urinary recovery of "unaltered" and "total" caronamide with mannitol. 2) The renal clearances of "unaltered" and "total" caronamide calculated on the basis of whole and ultrafiltered plasma. 3) A correlation of the extraction of "unaltered" and "total" caronamide and creatinine from the renal circulation with simultaneous renal clearances of the compounds on the basis of whole and ultrafiltered plasma.

Synthetic curare compounds. II. d-N-methyl-isochondrodendrine iodide and d-N-methyl-O-methyl-isochondrodendrine iodide. M. H. PELLETIER (by invitation) and DAVID FIELDING MARSH. *Department of Pharmacology, West Virginia University School of Medicine, Morgantown, W. Va.* d-Isochondrodendrine was isolated from an old sample of "Bebeerine Pure" and from the roots of Chondrodendron tomentosum. The curariform activity of its derivatives, d-N-methyl-isochondrodendrine iodide (iCM), and d-N-methyl-O-methyl-isochondrodendrine iodide (iCMMeO) was determined in comparison with the corresponding isomers, d-tubocurarine chloride pentahydrate (dT), and d-O-methyl-tubocurarine iodide trihydrate (dTMeO).

	dT	dTMeO	iCM	iCMMeO
Albino rats				
LD 50	0.27(0.22)*	0.032(0.022)	5.5(3.9)	3.0(2.2)
Rabbits				
Head drop 50	0.12(0.10)	0.016(0.011)	2.5(1.8)	0.6(0.4)
Holaday head drop	0.15(0.12)	0.020(0.014)	2.9(2.1)	0.7(0.5)
LD 50	0.35(0.28)	0.040(0.027)	4.5(3.2)	1.3(0.9)
Cat gastrocnemius muscle				
Equivalent paralysis	0.05(0.04)	0.007(0.005)	1.0(0.7)	0.2(0.14)

* All doses given in milligrams/kilogram body weight. Dose of equivalent contained curariform ion given in parenthesis.

Like the tubocurarine compounds, these isochondrodendrine derivatives have relatively little effect in intact animals other than lissive action on skeletal muscles.

Chronic intoxication of the CNS produced by the methadon side-chain. CARL C. PFEIFFER, ELIZABETH H. JENNEY (by invitation) and I. GERSH. *Depts. of Pharmacology and Pathology, Univ. of Illinois College of Medicine, Chicago 12.* The methadon side-chain, dimethyl, 2-chloropropylamine HCl, produces a non-lethal type of persistent ataxia when injected into mice or rats in a dosage range of 100 to 200 mgm./kgm. The LD-50 after intraperitoneal injection in the mouse is 234 ± 17 mgm./kgm., slope 7.4. The volatile compound dimethyl,2-chloropropylamine produces a similar syndrome in rats when concentrations as low as 40 parts per million are inhaled for 15 minutes. Brain specimens were collected from intoxicated rats after carotid perfusion with saline and 10 percent neutral formalin. Celloidin sections of cerebrum and cerebellum were made and stained with Toluidine Blue and Hematoxylin and eosin. Microscopic examination showed no changes in the cerebrum. Pathological changes in the cerebellum consisted of petechial hemorrhages in the gray and white matter, hyperchromatism and shrinkage of the Purkinje cells and mobilization of the glial cells. One or more of these lesions was present in all specimens examined. Similar CNS ataxias have been described after the administration of other chlorinated amines (Krop, S., et al: Fed. Proc., **6:** 347, 1947; A. Goldin et al: Bib. Sci. and Ind. Rept., **5:** 678, 1947) and also after chlorinated anesthetic agents (Abreu, B. E., et al: Anesth. and Analg., p. 131, May–June 1939). It is concluded that workers in chemical industry should avoid exposure to these volatile amines.

The nature of the prosthetic groups of analgesics and their possible action as blocking agents. CARL C. PFEIFFER, J. SANTOS-MARTINEZ (by invitation) and THEODORE R. SHERROD (by invitation). *Department of Pharmacology, University of Illinois College of Medicine, Chicago 12, Ill.* Studies on potent analgesic drugs, using Hirschfelder atomic models, suggest that analgesic drugs owe their pharmacological activity to three factors: 1) a methyl on tertiary nitrogen prosthetic group (quaternization does not enhance the analgesic action of methadon), 2) the presence of several active oxygen prosthetic groups at a distance of 7.0 to 9.0 Angstra from the methyl on nitrogen group, and 3) the presence of one or more

blocking moieties (phenyl, biphenyl, or dibutyl). The relationship of the prosthetic groups is independent of the general ring structure and obtains for the phenanthenes, biphenyls, piperidines, oxazolidinediones, etc. Since these prosthetic groups are similar to those of the acetylcholine-atropine series, compounds such as meperidine (demerol) which have a shorter distance between prosthetic groups than that postulated for the potent analgesic drugs, show decreased analgesic action and varying degrees of atropine-like action. Assuming that these analgesic drugs depress the CNS by blocking a specific CNS metabolite, the following known metabolites and their derivatives have been studied to determine if any will lower the normal pain threshold. The known amino acids studied are: glutamic acid, l-lysine, d-arginine, l-ornithine, and l-tryptophane. Synthetic derivatives have also been synthesized and their action will be reported.

Use of hyperglycemic response for estimating addiction potentialities of analgesic compounds. NILKANTH M. PHATAK, JAMES MALONEY (by invitation) and NORMAN DAVID. *Department of Pharmacology, University of Oregon Medical School, Portland, Oregon.* An objective evidence of morphine habituation was observed by Ro (Jap. J. Med. Sci. IV Pharmacology, **9**: 59, 1935). He noted that during development of tolerance to morphine in animals, hyperglycemic responses to successive injections become progressively less but upon withdrawal a hyperglycemic response appears at the expected time of injection. In a later biochemorphologic study of morphine derivatives, Emerson and Phatak (Univ. Calif. Publicat. in Pharm. **1**: 77, 1938) confirmed Ro's observations and suggested that the hyperglycemic withdrawal phenomenon might be useful for estimating addiction potentialities. The present study compares hyperglycemic responses to l-amidone (Merck), dl-amidone (Dolophine, Lilly), dl-isoamidone (Merck), meperidine (Demerol, Winthrop) and morphine in rabbits, using saline controls. Initial injections of all analgesic compounds produced hyperglycemia. The animals were injected daily for a period of six weeks, with weekly increments in dosage as follows: amidone compounds, 5, 6, 7, 8, 9 and 9 mg./kg.; morphine sulfate, 10, 20, 30, 40, 50 and 60 mg./kg.; meperidine HCl, 50, 75, 90, 100, 110 and 110 mg./kg. During development of tolerance greatest hyperglycemic responses followed injections of l-amidone and dl-amidone. During the withdrawal period the greatest hyperglycemic response occurred with l-amidone, was slightly less following morphine and dl-amidone, and negligible for dl-isoamidone and meperidine. Convulsions, opisthotonus and a cataleptic state with temporary respiratory arrest followed injection of the synthetic compounds. l-Amidone and dl-amidone produced these signs at the initia dosage of 5 mg./kg., while those noted for dl-isoamidone were less marked and did not appear until the dosage reached 8 mg./kg. Meperidine produced the same signs at 90 mg./kg.

Studies on the toxicology of podophyllotoxin and related substances. FREDERICK S. PHILIPS, MAYNARD B. CHENOWETH, and CARLTON C. HUNT (by invitation). *From the Departments of Pharmacology, Cornell Univ. Medical College and the Sloan-Kettering Institute for Cancer Research, New York, N. Y.* Recent demonstrations of colchicine-like effects of podophyllotoxin and podophyllin in arresting cell-mitosis in metaphase and actions of the drugs against *condylomata acuminata* and experimental tumors stimulated studies of their toxicology. Intravenous LD_{50}'s of podophyllotoxin were: rats, 8.7, cats, 1.7, rabbits, ca. 5 mg./kg.; intraperitoneally: mice, 33, rats, 15 mg./kg.; intramuscularly: cats, ca. 4, rats, ca. 3 mg./kg. Cats and dogs receiving seven or more daily, intravenous doses of 0.5 mg./kg. of podophyllotoxin survived without alarming consequences. Similarly mice tolerated 8 mg./kg. daily intraperitoneally. In confirmation of previous investigations podophyllin was found to contain toxic components not identical with podophyllotoxin. Several residues remaining after solvent extraction of podophyllotoxin from podophyllin proved to be equally or even more toxic in rats and mice than podophyllotoxin itself. Animals receiving fatal doses of podophyllotoxin usually succumbed within 24 hours. Initial signs included emesis, defecation with tenesmus, and respiratory stimulation. Dogs occasionally developed bradycardia and ventricular extrasystoles (reversed by atropine). Muscular weakness, heralded by hind-leg ataxia, resulted in eventual prostration. Terminally respiration was slow and labored, blood pressure fell to shock levels, and death followed respiratory failure. In cats and rats, but not in dogs, fatal doses often caused severe pulmonary damage. A small proportion of animals receiving minimum lethal doses exhibited progressive cachexia and succumbed after several days. These fatalities were not associated with gastro-intestinal, hepatic, or renal damage or disturbances of hematopoiesis. However, toxic doses did elicit a marked granulocytopenia within 1 hour after administration from which animals recovered promptly.

Spasmolytic and other pharmacologic properties of β-piperidinoethyl phenyl-α-thienylglycolate. RAYMOND W. PICKERING and RICHARD C. BURNETT (introduced by BENEDICT E. ABREU). *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco.* β-piperidinoethyl phenyl-α-thienyl glycolate (P.P.T.) in doses ranging from 0.4 mgm./kg. to 8 mgm./kg., subcutaneously produced spas-

molysis, lasting from 1 to 62 minutes in 18 monkeys whose colons had been rendered hyperactive with morphine. Trasentin in doses as high as 10 mgm./kg., subcutaneously did not produce comparable effects. In preliminary experiments on anesthetized dogs, P.P.T. 2.5 mgm./kg. I.V., depressed the activity of the ileum and jejunum for 122 minutes. P.P.T. usually produced spasm in the non-pregnant guinea pig uterine strip in concentration of 5×10^{-3} mgm./ml. occasionally relaxation was observed at this concentration. In 33 human subjects, P.P.T. did not produce any untoward effects in doses ranging from 2.5 mgm. to 35 mgm. given orally. One subject who received 35 mgm. complained of vertigo, drowsiness, dimness of vision and chills for 40 minutes after ingestion. Complete recovery occurred within 60 minutes. In other subjects lower doses did not produce: hypnotic or analgetic effects, central nervous system stimulation, cycloplegia, mydriasis, xerostomia, nor changes in respiration, blood pressure, electrocardiographic tracings nor cardiac rate. Examination of urine revealed no abnormalities. In 4 subjects given 5 and 10 mgm. sublingual doses of P.P.T., spasmolysis for an average of 36 minutes (30–62 minutes) occurred. There was no increase in gastric acidity upon sublingual administration of P.P.T. to 4 individuals.

Abolition of extra systoles in the perfused isolated rabbit heart by procaine and monocaine hydrochlorides. HARRY J. PRATT (introduced by R. BEUTNER). *Department of Pharmacology, Hahnemann Medical College, Phila., Pa.* In connection with the study of the cardiac toxicity of Procaine and Monocaine Hydrochlorides on the perfused isolated rabbit heart, it was observed that extra systoles were always shown by the normal rabbit heart during the control period. Following the addition of Procaine or Monocaine in a concentration of 1:100,000 to the perfusion fluid, complete abolition of extra systoles was observed in 100% of the experiments. Monocaine Hydrochloride in a concentration of 1:150,000 in the perfusion fluid prevented the continuance of extra systoles in about 50% of further experiments and at a concentration of 1:500,000, no effect upon the prevention of extra systoles was observed. Lower concentrations of Procaine Hydrochloride were not used. These findings are in agreement with the clinical use of intravenous administration of local anesthestics in cardiac irregularities elicited by Cyclopropane anesthesia or from other causes, since they demonstrate the regularizing influence of local anesthestics upon the heart.

Toxicological properties of the causative agent of canine hysteria. JACK L. RADOMSKI, GOEFFREY WOODARD, CARTER D. JOHNSTON (introduced by ARNOLD J. LEHMAN). *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington 25, D. C.* Recent work in this and other laboratories has established that a reaction product of nitrogen trichloride (NCl_3) with the protein of wheat flour is the etiological agent of canine hysteria. The cumulative characteristics of this toxic agent have been investigated. Several groups of 10 month old dogs were fed NCl_3-treated gluten (6 gm. NCl_3/kg. gluten) in doses ranging from 2.5 to 0.1 gm. per kg. per day. When the average number of days required for the animals to come down with running fits on each dosage level was plotted against the daily dosage, a hyperbola-like curve was obtained. This curve becomes asymptotic at about 0.1 gm./kg./day. Apparently the dog is capable of destroying and/or excreting the amount of toxic principle contained in about this amount of NCl_3-treated gluten. Translating these data into other terms, it means that a dog could consume normally treated flour (1.5 gm. of agene/100 lbs.) to the extent of ¼ of his diet without developing toxic effects. The sensitivity to constant dosages of treated gluten per kilogram of body weight varies with the age of the dog, 12 month old dogs being more sensitive than 6 month old dogs. Differences in sex susceptibility have not been observed. Various drugs including those used in epilepsy have been tested for their value in canine hysteria.

The effect of cyclopropane on the work capacity of the dog heart. BARBARA RENNICK (by invitation), S. DONALD MALTON (by invitation), and GORDON K. MOE. *From the Department of Pharmacology, the University of Michigan, Ann Arbor.* It has been established that the cardiac output of animals under cyclopropane anesthesia is maintained at normal levels until concentrations producing respiratory arrest are reached. It has been assumed that in anesthetic concentrations the gas produces no impairment of cardiac function. It is, however, impossible to estimate the effect of a drug on cardiac reserve by measurement of cardiac output alone, since this may be maintained within relatively normal limits in the presence of severe limitation of cardiac work capacity. In heart-lung preparations exposed to various concentrations of cyclopropane increased work loads were offered by increasing arterial resistance; responses were measured in terms of auricular pressures and cardiac output. In intact animals the work load was increased by elevating arterial pressure with the aid of a pressure regulator attached to the abdominal aorta, and the responses were estimated in terms of right auricular pressure changes. After the responses to graded pressure loading were measured during exposure to various concentrations of cyclopropane, sodium pentobarbital anesthesia was substituted. Artificial respiration was maintained throughout. The work capacity of the isolated heart was significantly reduced by 20% cyclo-

propane, and greatly reduced by 40%. In the intact animal the elevation of venous pressure in response to pressure loading was significantly greater under 20% cyclopropane than with 30 mgm/kg. of pentobarbital, and markedly greater under 40% cyclopropane than with 45 mgm/kg. of pentobarbital. We conclude that cyclopropane in anesthetic concentrations diminishes the work capacity of the dog heart.

Detoxification of 2-benzyl-imidazoline hydrochloride (Priscol) by incubation with hepatic slices from the rat. B. RICHARDS (by invitation), A. CAMERON (by invitation), B. CRAVER, E. HERROLD (by invitation). *Division of Pharmacology, Research Department, Ciba Pharmaceutical Products, Inc., Summit, New Jersey.* Two methods were employed to determine whether 2-Benzyl-Imidazoline Hydrochloride was detoxified "in vitro" by incubation with hepatic slices from the rat. An investigation of the toxicity of various imidazolines upon isolated tissues first led to these studies of detoxification. By means of the Warburg technique, it was found that a concentration of 10 mg/ml. of Priscol caused little or no inhibition of the respiratory rate of hepatic slices from the rat but a considerable decrease in the respiration of renal slices from the rat. The technique of Finklestein, as described elsewhere in the Proceedings, was employed with an extension to include non-cardiac tissues. A dose of 10 mg/ml. of Priscol, after incubation with hepatic slices from the rat, proved less toxic to renal slices from the rat. We interpreted this to indicate hepatic detoxification. In the second method the drug was incubated with hepatic slices from the rat and the Priscol subsequently bioassayed on the isolated, perfused heart of the guinea pig. Priscol in small doses exerts a positive inotropic action on this heart. Priscol, first incubated with liver, is much less active in its cardiac effect than Priscol not so incubated. The minimally toxic doses used in these experiments far exceeded any level that would be attained therapeutically. These experiments confirm those of others indicating the feasibility of this approach to detoxification.

Further analysis of the influence of autonomic innervation on drug responses of the heart and gut in unanesthetised dogs. JAMES A. RICHARDSON (by invitation) and R. P. WALTON. *Dept. of Pharmacology, Medical College of South Carolina.* Heart slowing in dogs, as produced by morphine (1.0 mgm. per kgm. subcutaneously) was not present during the few days survival interval after bilateral cervical vagotomy. Morphine-induced gut spasm as determined by Thierry fistula tracings was, however, obtained during a 36 hour interval after cervical vagotomy and during a 6 weeks interval after supra-diaphragmatic vagotomy. (Morphine test doses were either 0.5 or 1.0 mgm. per kgm. subcutaneously.) Also following vagotomy at the diaphragm level, acetylcholine effects on gut movements were essentially unchanged in some cases and moderately intensified in others. (Acetylcholine administration before and after vagotomy was effected by intravenous infusion at a uniform rate of 0.2 mgm. per kgm. per minute for 5 minutes.) High spinal anesthesia with procaine had a consistently depressant effect on the typical Thierry fistula tracing. Dibenamine in doses up to 10 mgm. per kgm. intravenously either failed to affect the character of the typical Thierry fistula tracing or produced moderate increase in general tone level; following these dibenamine injections, characteristic epinephrine depressant effects and characteristic morphine stimulant effects were obtained.

Elevation of serum protein-bound iodine after large doses of radio-active iodine. DOUGLAS S. RIGGS (introduced by OTTO KRAYER). *Department of Pharmacology, Harvard Medical School and The Thyroid Clinic, Massachusetts General Hospital.* Shortly after the administration of therapeutic doses of radio-active iodine to hyperthyroid patients, a transient increase in metabolic rate, swelling and tenderness of the thyroid, and radiation sickness have occasionally been noted. None of these reactions has been alarming. However, Salter has described a marked elevation of plasma protein-bound iodine and the development of hyperthyroidism when a large dose of radio-active iodine was given to a patient with massive metastases from a functional carcinoma of the thyroid gland. Additional instances of such elevations are reported here. The protein-bound iodine of a patient with thyroid carcinoma rose from 4.3 to 15.0 micrograms per cent 7 days after a "thyroidectomizing" dose of I^{131}. A significant, though smaller, increase was also observed in another patient with thyroid carcinoma and mild hypothyroidism. These two patients were in the Sloan Kettering Institute. The protein-bound iodine of a patient with Graves' disease rose from 16.2 to 23.0 micrograms per cent 7 days after a therapeutic dose of I^{131}. A similar increase occurred in a patient with severe hyperthyroidism, but it could not be ascribed conclusively to therapy with radioactive iodine. This patient died of coronary occlusion 19 days after treatment. The transient rise in serum protein-bound iodine is presumably due to a sudden release of preformed hormone from the gland to the blood stream when the thyroid is exposed to intense radiation. These observations suggest that large doses of radioactive iodine may precipitate an acute exacerbation of hyperthyroidism in certain patients.

The effect of globin insulin and protamine zinc insulin on the diurnal blood sugar curve and the daily glycosuria of diabetics, and the clinical use

of globin insulin. JOSEPH THOMAS ROBERTS. *From The University of Arkansas School of Medicine, Little Rock, Arkansas, and Gallinger Municipal Hospital, Washington, D. C.* Globin insulin controlled the average blood sugar curve and daily glycosuria of 97 diabetics in a more nearly normal way than did the same doses of protamine zinc insulin. With globin insulin, the three preprandial and midnight blood sugar levels had normal average values, while with protamine zinc insulin in equal doses only the fasting a.m. value was normal. With globin insulin, the three postprandial rises in blood sugar levels were smaller than with the other preparation. Globin insulin was effective in maintaining good control of diabetics in the out-patient clinic, and seemed of some value in smoothing the recovery curve of blood sugar values of diabetic acidosis when used in conjunction with regular (crystaline) insulin. Local and systemic reactions were fewer and milder with globin insulin than with protamine zinc insulin. Serious reactions with globin insulin may follow ignoring the precautions generally used with protamine zinc insulin.

Comparative intragastric and local toxicities of Tert-butyl-hydrogen peroxide, di-tert-butyl-peroxide, and 2,2 bis-di-tert-butyl-peroxybutane. R. R. ROLLINS (introduced by HAMILTON H. ANDERSON). *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco.* Tert-butyl hydrogen peroxide has been reported to have biologic activity (Mendex, R.; Jour. Pharmacol., **82**: 377, 1944). It is compared here to two of its derivatives and to ascaridol. The peroxides were administered to rats and mice intragastrically, either undiluted or in cottonseed oil (see table). The percentage purity of the compounds is indicated. The lower boiling

Compound	Approx. purity	No. of rats	No. of mice	Estimated $L.D._{50}$, ml./kg.		Time to kill 50% of mice (sat. vapor-air mixture)
				Rats	Mice	
Ascaridol.....	85%	22	12	0.20	0.40	Undetermined
tert-butyl hydrogen peroxide (*TBHP*).....	60%	29	14	1.5	2.0	10–15 minutes
di-tet-butyl peroxide (*TBP*)...	90%	17	12	17.5	20.0	10–15 minutes
2,2 bis-di-tert-butyl peroxybutane (*TBPB*)	80%	24	36	12.5	17.5	more than 510 minutes

components of 2,2 bis compound (TBPB) were removed upon aeration. The differences in toxicity on inhalation are probably related to differences in boiling points. The more volatile TBHP and TBP killed in less time because of higher vapor concentrations. The tert-butyl hydrogen peroxide was the most irritant of the three compounds when applied topically. In one individual, dermatitis venenata developed after brief contact. 2,2 bis-di-tert-butyl peroxybutane had a similar action on the skin and eye of the rabbit as butyl acetate when compared with previously reported results (McOmie and Anderson—to be published).

Preliminary studies of ascaricidal activity (*in vitro*) indicated that only tert-butyl hydrogen peroxide had an activity of the same order as ascaridol.

Adenylpyrophosphatases and other phosphatases in the cell surface of living yeast. ASER ROTHSTEIN (by invitation) and REBECCA MEIER (by invitation) (introduced by HARVEY B. HAAG). Various phosphate compounds such as adenyltriphosphate (ATP), adenyldiphosphate (ADP), Na glycerophosphate, inorganic triphosphate, or inorganic pyrophosphate, added to a suspension of washed, starved living Bakers' yeast (Standard Brands) are hydrolyzed with the production of inorganic ortho-phosphate. An extensive study of muscle and yeast ATP breakdown indicates that during hydrolysis, the products (adenylic acid and inorganic phosphate in the ratio of 1:2) and unused substrate can be recovered quantitatively from the medium. In addition, if ATP containing radioactive phosphate (P_{32}) is hydrolyzed by the cells, all of the P_{32} is recovered in the medium with none in the cells. These experiments indicate that the enzyme is at or in the cell surface. However, the enzyme is firmly bound, because the medium separated from the cells shows no activity. A study of the pH-activity curves and of competition or non-competition of various substrates with each other, indicates that ATP, ADP, and inorganic triphosphate are hydrolyzed by an enzyme with a pH optimum of 3.5 and almost zero activity at 7.0 and higher. The Na glycerophosphate and inorganic pyrophosphate are hydrolyzed at a lower rate by other enzymes which are active at pH 6.0 and below but with no well defined optimum. The limiting substrate concentration for the ATP hydrolysis is about 0.7×10^{-3} M. With optimum ATP concentrations the rate of hydrolysis is directly proportional to yeast concentration, indicating a first order reaction. The time course of hydrolysis, however, follows an asymptotic curve rather than a linear function due not to destruction of enzyme, but apparently to inhibition by the products of the reaction, inorganic phosphate and adenylic acid. A synergistic effect between these two substances seems to be involved. The inhibition due to the phosphate is greatly increased by the adenylic acid, which by itself has no effect.

Effect of quinidine on the metabolism of the rat heart. PAUL R. SAUNDERS (by invitation), J. LEYDEN WEBB (by invitation), and CLINTON H. THIENES. *Department of Pharmacology and Toxicology, School of Medicine, University of Southern California, Los Angeles, California.* Quinidine at a concentration of 0.0003M was found to depress the endogenous oxygen-uptake of rat ventricle slices approximately 50 per cent, and also to decrease the ability of glucose to maintain the oxygen-uptake of such slices. This concentration has been found to produce typical quinidine effects on the isolated perfused rat heart. Only slight effects were observed at this concentration on the utilization of succinate, lactate, pyruvate, and malate, either with tissue slices or homogenates, although higher concentrations were somewhat inhibitory to several dehydrogenation reactions. Further evidence has indicated that quinidine exerts its depressant action on rat heart metabolism probably chiefly by inhibition of the earlier stages of glucose breakdown. The effects of quinidine on the breakdown and synthesis of high-energy phosphate bonds in rat heart homogenates has also been investigated. It apparently does not markedly affect adenosinetriphosphate breakdown at several low concentrations which depress the isolated perfused heart.

Use of oxygen consumption method for measuring antithyroid activity of n-propylthiouracil, thiouracil and 2-aminothiazole in rats. EDWARD SAXEY (by invitation) and NILKANTH M. PHATAK. *Department of Pharmacology, University of Oregon Medical School, Portland, Oregon.* Direct measurement of the O_2-consumption rate seems logical in preference to indirect methods now utilized to estimate the relative effectiveness of antithyroid drugs. Using a modified Peoples' Metabolar (Phatak and Saxey, J. Amer. Pharm. Assoc., **36**: 105, April, 1947) we have determined weekly O_2-consumption rates in rats before and after treatment with thiouracil, n-propylthiouracil and 2-aminothiazole. The latter compound was fed in drinking water as a 0.1% solution while the thiouracils were injected intraperitoneally in doses of 100 mg./kg. per day. The duration of treatment varied from two to four weeks. After administration of n-propylthiouracil the O_2-consumption rate of rats showed a decrease from a mean value of 20.4 cc./min./kg. bodyweight to 19.0 cc./min./kg. at the end of two weeks. Within a comparable period the O_2-consumption rate of the rats given 2-aminothiazole decreased from a normal value of 19.3 cc./min./kg. to 17.6 cc./min./kg. with a further decrease to 16.3 cc./min./kg. at the end of four weeks. Thiouracil at 100 mg./kg. dose gave no indication of effectiveness after two weeks of administration.

The circulation of penicillin in the lymph. R. J. SCHACHTER (introduced by N. ERCOLI). *Dept. Pharmacology and Chemotherapy, Warner Inst. Therap. Research, New York City.* From the work of Romansky) New England J. Med. **223**: 577: 1945) it is known that penicillin might still be present in the urine after the blood stream apparently is penicillin-free. We found, following an intramuscular injection of a suspension of penicillin in oil with vasoconstrictor, that the duration of penicillin in the tissues is more than twice as long as in the blood stream. An explanation of these observations indicated a follow-up of the fate and circulation of penicillin in the body, particularly at the end period of the blood level curve and afterwards. This investigation revealed that the circulation of penicillin in the lymph might be as important a factor in its transport as the blood circulation. As a rule, the lymph collected from the cannulated thoracic duct of dogs contained detectable amounts of penicillin for significantly longer periods than the blood stream. The penicillin present in the lymph—probably collected from the organs—eventually reaches the blood stream where it is diluted to a point beyond detection, then again concentrated in the kidney and eliminated. This explains why the blood apparently is free from penicillin while the kidneys still excrete detectable amounts. The present findings on the prolonged lymphatic circulation, the persistence of penicillin in the organs, and the chemotherapeutic activity without detectable blood levels, observed by Ercoli, Lewis, Schwartz, and whitehead (this issue), lead to the conclusion that the *blood level* is only one of many factors related to the effectiveness of penicillin therapy.

Malaria chemotherapy: 2. The response of sporozoite-induced infections with Plasmodium cynomolgi to various antimalarial drugs. L. H. SCHMIDT, ROCHELLE FRADKIN (by invitation), WANDA SQUIRES (by invitation), and CLARA S. GENTHER (by invitation). *Christ Hospital Institute of Medical Research, Cincinnati, Ohio.* The activities of various antimalarial drugs against trophozoite-induced infections with *Plasmodium cynomolgi* were described in the preceding paper (Fed. Proceedings, this issue, p. 221). This report deals with response of sporozoite-induced infections to quinine, quinacrine, chloroquine, oxychloroquine, chlorguanide (*Paludrine*), pamaquine, pamaquine plus quinine, pentaquine and pentaquine plus quinine. Approximately 250 rhesus monkeys were infected intravenously with measured doses of sporozoites. Treatment with one of the above drugs was initiated either in the ascending phase of the primary attack, or during the first, second or third relapses in cases where infections were not cured by previous medication. In all cases drugs were administered once daily, the duration of treatment being either 7 or 14 days. Criteria of cure were identical with those used in the tropho-

zoite-induced infections (loc. cit.). Results were as follows. Quinine, quinacrine, chloroquine, oxychloroquine, and chlorguanide suppressed the clinical attacks of the sporozoite-induced infections in the same doses as were effective against the trophozoite-induced disease. Cures were not effected by these drugs, however, even when doses were administered which were 60 times greater than those which cured trophozoite-induced infections. In contrast, pamaquine and pentaquine, administered either alone or with quinine did effect cures. Dose for dose these 8-aminoquinolines appeared to be equally effective. These results indicate rather clearly that the chemotherapeutic characteristics of *P. cynomolgi* infections in the rhesus monkey are identical with those of *P. vivax* infections in man.

Comparison of therapeutic values of several antimonials in experimental schistosomiasis mansoni in mice. MAXWELL SCHUBERT and EVELYN GOLDBERG (by invitation). *From the Department of Therapeutics, New York University College of Medicine.* Methods previously described for production of standard infections of Schistosomiasis mansoni in mice, for systematic autopsy of these mice and estimate of the effects of drugs on such infections, had been applied only to rough screening of drugs. They have now been used in an attempt to measure therapeutic effectiveness of several antimonials on the disease in mice. Two methods are discussed. The first is the simple therapeutic index, the ratio of the chronic LD50 to the CD50 where the latter quantity is the dose that frees fifty per cent of the mice completely of worms. The second is based on the fraction of worms destroyed. Because of the high variability of worms found per mouse even in control groups this fraction is taken as nine tenths rather than one half. So the second measure of therapeutic effectiveness is the ratio of the LD50 to the dose that reduces the average worm count to about a tenth of its value in control untreated mice. Errors in both of these indexes are discussed in detail. Within the wide limits of error to which the values being measured are subject, both indexes have similar magnitudes. For sodium antimony tartrate, ureastibamine, neostibosan and neostam the indexes are below one. Fuadin is the best of the antimonials currently used, its index is one. Two oil soluble antimonials are the only drugs known to have an index greater than one. Implications are discussed.

The chemotherapeutic action of a number of furan derivatives. LLOYD D. SEAGER, *Department of Pharmacology, The Woman's Medical College of Penna.* Twenty furan derivatives were studied for their chemotherapeutic effectiveness against Trypanosoma equiperdum infections. Mice were inoculated intraperitoneally with 500,000 to 1,000,000 parasites/kg. With the strain of Equiperdum used this produces 100% fatality in untreated mice within 5 days. Drugs were given by stomach tube 3 times daily for 4 days. Soluble compounds were tested also by the single intraperitoneal method. Furfural, 5-nitro-2 furaldehyde semicarbazone, furylacetate, furfuryl alcohol, furoic acid and durilacrylic acid were found to be active. Additive effects but no synergism was found between the active furan derivatives. Furfural was found to have additive effects but no synergism with acriflavine, stilbamidine and pentamidine. None of the furan compounds tested were found to influence the development of T. cruzi infections in mice.

The nutritive value of the fatty acids of lard esterified with a polyethylene glycol. C. BOYD SHAFFER and FRANCES H. CRITCHFIELD (introduced by PAUL L. MCLAIN). *Chemical Hygiene Fellowship, Mellon Institute, Pittsburgh, Pennsylvania.* The increasing use of the polyglycol esters as pharmaceutical vehicles and as an additive in bread to prevent the retrogradation of starch renders important studies of their biochemical behavior. The present report gives information on the nutritive value of the esters obtained by combining the fatty acids present in lard with a polyethylene glycol of average molecular weight 400. The plan of the experiment is similar to those described by Lepkovsky, Ouer and Evans (J. Biol. Chem. **108:** 431, 1935). Lard fatty acids were prepared according to the preceding reference. Part of the product was esterified with polyethylene glycol 400 to prepare a monoester, while the remainder was resynthesized to the triglycerides. Two groups of twenty-four weanling rats each were fed a diet consisting of sucrose 37.5, casein 32.5, salts 4, and cellulose 5. In group I (controls) fat was supplied as 21 parts of the resynthesized glycerides, while in group II it consisted of 21 parts of the polyglycol ester. Purified vitamins were supplied as daily supplements. At the conclusion of 47 days feeding the mean weight gain of the polyglycol ester-fed group (group II) was 110 gms. per rat as opposed to 104 gms. in the controls, while the daily food consumption in group II was 10 gms. per rat as compared with 8.5 gms. in group I. Neither of these differences is statistically significant, nor were the growth curves significantly different. No evidence of gross pathology was observed in either group at sacrifice.

Comparative effects of d-desoxyephedrine and the isomers of amphetamine on smooth muscle. FRED SHAFFER (by invitation), P. L. EWING and G. A. EMERSON. *Department of Pharmacology, University of Texas Medical Branch, Galveston, Texas.* Quantitative comparisons of the effects of d-desoxyephedrine and d-, l-, and dl-amphetamine on intact and isolated smooth muscle of animals are made. The property of evincing the phenomenon of tachyphylaxis is confirmed for these compounds. On the rat intestine, the inhibitory action of epinephrine is antagonized by these phenalkylamines

and is expressed quantitatively as the pA. Other antagonisms are reported which help elucidate the mechanisms of action involved.

Onset and duration of action of quinidine in the heart after oral administration in man. S. J. SHANE (by invitation), CONRADO DAYRIT (by invitation), JOSEPH G. BENTON (by invitation), ELAINE W. COTLOVE (by invitation), LAWRENCE W. HANLON (by invitation), WALTER MODELL, and HARRY GOLD. *Department of Pharmacology of Cornell University Medical College, Cardiovascular Research Unit of the Beth Israel Hospital, and Cardiac Service of the Hospital for Joint Diseases, New York.* The effect of quinidine sulfate was determined by the change in the speed of the circus movement of the auricle in patients with auricular fibrillation. Each patient received a series of doses, varying from 0.1 to 1.2 gm. at one time, the different doses being given at intervals of several days. After a control electrocardiogram was taken for the measurement of the F–F intervals, additional tracings following the oral dose of the drug were taken at intervals of 2 hours. In a group of 10 patients, the results show that the maximum effect is already present in 2 hours in the vast majority of cases after doses of from 0.1 to 1.2 gm. The effect wears off gradually at speeds varying with the dose. It has completely disappeared in 8 hours after 0.1 gm. and three-fourths of the effect disappears in 8 hours after 1.2 gm.

Methadon derivatives of pharmacological interest. THEODORE R. SHERROD (by invitation), ROBERT KAISER (by invitation), J. SANTOS-MARTINEZ (by invitation) and CARL C. PFEIFFER. *Department of Pharmacology, University of Illinois College of Medicine, Chicago 12.* From the pharmacological viewpoint it is hoped that at least five types of methadon derivatives may be synthesized: 1) a short-acting nondepressant analgesic for use in obstetrics, 2) a long-acting orally-effective analgesic to obtund the pain of cancer, 3) a potent spasmolytic-analgesic compound, 4) a narcotic analgesic with a wide margin of safety for preanesthetic medication, and 5) a potent, persistent analgesic which perhaps, by chemical lobotomy, may permanently lower the pain threshold in causalgic states. With these concepts in mind the following compounds are of interest:

	Therapeutic index
1) Methadon HCl	2.3
2) Isomethadon HCl	3.1
3) N,N dimethyl-3,3-diphenyl-4-imino-2-methylhexylamine HCl	4.0
4) γ dimethylamino-α, α diphenylvaleric acid HCl	8.0
5) 1-dimethylamino-2-methyl-3, 3-diphenyl-4-acetoxyheptane HCl	11
6) 2 dimethylamino-4, 4-diphenyl-5-hexylidenacetylimine HCl	12
7) 2-dimethylamino-4, 4-diphenyl-5-acetoxyheptane HCl	14
8) 2-morpholino-4, 4-diphenyl-5-acetoxyheptane HCl	40

Compound No. 4 is unique in this series because of an atropine-like effect on the intestine of the anesthetized dog. The other derivatives have a spasmogenic effect on the intestine similar to that of methadon. Compounds No. 5 to 8 are of interest because of their high therapeutic indices and long duration of analgesic action. The pharmacology of these compounds will be discussed.

Blood levels of thiopental (Pentothal) following repeated intravenous administration to the dog. F. E. SHIDEMAN, A. R. KELLY (by invitation), and B. J. ADAMS (by invitation). *Department of Pharmacology, University of Michigan.* The method of Jailer and Goldbaum was employed to follow blood levels of thiopental in dogs receiving repeated doses of the drug. One group of dogs was used to study the plasma levels of thiopental when moderate doses (10 mg./kg.) were repeated at short intervals (after complete recovery from narcotic action). Under these circumstances, the following evidence suggests that some acute tolerance to the depressant action of this drug is developed: a. The plasma level at which the righting reflexes return is higher with each successive dose even though the actual duration of narcosis progressively increases. b. A significant concentration of the drug is still present in plasma even after complete objective recovery, and this amount increases with each successive dose. Preliminary experiments were done in a second group of dogs to study the plasma levels and decay curve following the single daily administration of a larger dose (20 mg./kg.) of thiopental for 7–9 days. The similarity of the decay curve on the first and last day suggests, but does not prove conclusively, that under these circumstances neither tolerance nor cumulation occurs.

The effect of hepatic dysfunction in man on the duration of action of thiopental (pentothal). F. E. SHIDEMAN, A. R. KELLY (by invitation), L. E. LEE (by invitation), V. F. LOWELL (by invitation) and B. J. ADAMS (by invitation). *Department of Pharmacology, University of Michigan and Ypsilanti State Hospital, Ypsilanti, Michigan.* It has been previously demonstrated that the liver is the major organ involved in the detoxication of Pentothal by the rat (Fed. Proc. **6**, 344, 1947). The following experiments were performed to determine if such were also the case in man. Bromsulfalein retention and serum proteins were determined in individuals with and without a history of liver disease. Nine patients with a normal bromsulfalein retention and A/G ratio (group A—normal liver function) and six with an abnormally high bromsulfalein retention and an abnormally low A/G ratio (group B—reduced liver function) were selected for study. Each patient received 4 mg/kg. of Pentothal Sodium in a 4 per cent solution intravenously, injected over a period of 30 seconds. Duration of action was determined as the interval

elapsing between the administration of the drug and the time when the individual could stand without support. The significant mean duration of action for group A was 149.4 seconds (t = 17.62) and for group B was 488.5 seconds (t = 4.72). A significant increase in the duration of action of Pentothal was thus demonstrated in those patients with impaired liver function (t of the difference of means = 3.32).

The specificity of the determination of alcohol in biological fluids. H. WARD SMITH (introduced by G. H. W. LUCAS). *Department of Pharmacology, University of Toronto.* Several high blood values for "apparent" alcohol in cases of diabetic acidosis have been reported in the literature. A reinvestigation was made of the possible values for "apparent" alcohol which appeared in advanced diabetic acidosis in experimental animals and in humans; measurements were made on blood and urine. In the course of this study, it was found necessary to specify completely the conditions for collection and treatment of sample, in order to minimize errors due to contaminants other than ketones. A "specific" method for alcohol was developed which was based on the Widmark desiccation procedure and which used a pretreatment of blood or urine samples by a modification of the Friedmann-Klaas reagents for saliva. The excess dichromate was determined colorimetrically by the use of the chromatediphenylcarbazide complex. When conditions for oxidation were completely specified, a theoretical recovery of alcohol added to blood or urine was obtained, using 0.5 cc. samples with a precision of ±2% over a wide range of concentrations. It is concluded that conditions can be chosen for the chromic acid oxidation such that the amounts of acetone possible in blood are of no practical significance.

Method for kymographic recording of CSF pressure; effects of histamine, antistine, PBZ, et al., thereon. J. SMITH (by invitation), A. CAMERON (by invitation), B. CRAVER' *Division of Pharmacology, Research Department, Ciba Pharmaceutical Products, Inc., Summit, New Jersey.* The effects of histamine upon canine spinal fluid pressure and the antagonistic actions exerted against them by the two antihistaminics: 2-(N-Phenyl-N-benzylaminomethyl)-imidazoline Hydrochloride (Antistine) and N,N-Dimethyl-N′-benzyl-N′(a-pyridyl)-ethylenediamine Monohydrochloride (Pyribenzamine) were investigated. A method was devised for the continuous kymographic recording of cerebral spinal fluid pressure, since none was available. A blunt 20 gauge needle was inserted into the cisterna magna and the changes in pressure transmitted to a sensitive water manometer. The effects of histamine were quite variable: increases, decreases, but more commonly bi-phasic reactions with an initial fall and a secondary rise. The antihistaminics antagonized about equally histamine's action on the CSF pressure and the blood pressure. The effects upon the pressure of a series of standard drugs were also investigated and the conclusion drawn that the effects upon the CSF pressure usually paralleled the effects upon the blood pressure. In general the results suggested that this method was an indirect measure of cerebral blood flow; but this hypothesis could be tested only by concomitant direct measures of the flow. The antihistaminics themselves usually caused a decrease or no change in the CSF pressure so that their clinical use probably would not be contraindicated in conditions in which that pressure was abnormally increased.

The tissue distribution and toxicity of emetine. PAUL K. SMITH, ABRAHAM I. GIMBLE (by invitation) and CLARKE DAVISON (by invitation). *From the Department of Pharmacology, The George Washington University School of Medicine, Washington, D. C.* Methods were investigated for the determination of the distribution of emetine in animal tissues. Results, using the methyl orange technique described by Brodie and Udenfriend (J. Biol. Chem. **158**, 705–14, (1945)) and a method utilizing the ultra-violet absorption bands, were in satisfactory agreement. Two hours after intraperitoneal injection of the drug in rats, the concentrations were highest in the liver with appreciable amounts in the lung, kidney and spleen. The concentrations in muscle, brain and blood were very low. In spite of the high cardiac toxicity of this substance, the concentrations in the heart were also low. After 48 hours, the concentrations in the liver, lung and kidney were approximately half the concentrations found at the end of two hours. Samples of the whole mouse were analyzed at intervals up to 35 days after intraperitoneal administration of the drug. The amount of emetine recovered decreased slowly until approximately the eighth day and remained approximately constant for at least 35 days with doses varying from 40 to 80 mgm. per kgm. The amount recovered after eight days was fairly constant; that is, there had apparently been a greater excretion or destruction of the drug after high doses than after low doses. The intraperitoneal LD_{50} in rats was approximately 17 mgm. per kgm. and in mice 62 mgm. per kgm. over a seven day period.

The distribution and excretion of antimony following administration of antimony potassium tartrate and neostibosan. RALPH G. SMITH, JAMES Y. P. CHEN (by invitation) and MARGUERITE MAGEE (by invitation). *Department of Pharmacology, Tulane University School of Medicine, New Orleans.* Antimony potassium tartrate (10 mg./kg.) and Neostibosan (80 mg./kg.) representing tervalent and quinquevalent antimony compounds were injected intraperitoneally in single doses into hamsters. Series of animals were sacrificed at 1, 6,

12, 24 or 48 hours after injection. Blood, heart, lungs, spleen, liver, muscle, bone, kidney, bile, gastrointestinal tract and contents, urine and feces were analyzed for antimony by the colorimetric method of Maren. With antimony potassium tartrate a decrease in antimony concentration in tissues and blood usually occurred after 6 to 12 hours but the content 48 hours after administration was at least 50 per cent of that at the one hour period, with the exception of kidney, gastrointestinal tract and bile which showed still lower values. The average concentrations in heart, spleen, liver, bone and gastrointestinal tract were higher at the 6 or 12 hour periods than 1 hour after injection but the differences involved were not statistically significant. With Neostibosan the decrease in antimony concentration over the 48 hour period was more rapid falling to from 20 to 50 per cent of the one hour values. Kidney and liver on the contrary showed maintained or increasing concentrations throughout. In comparison with other tissues, high concentrations of antimony occurred in liver, bile and somewhat in spleen after antimony potassium tartrate and in spleen, kidney, liver and bile after Neostibosan. In accord with previous reports more antimony was excreted by the bowel than by the kidney after antimony potassium tartrate, the reverse occurring after Neostibosan.

Initial studies of the inhalation toxicity of beryllium sulfate. GEORGE F. SPRAGUE (by invitation), ALTON G. PETTENGILL (by invitation), and HERBERT E. STOKINGER (by invitation). Introduced by HARVEY B. HAAG. Data from 56 animals representing 6 species were collected to supply information on the toxic effects caused by the inhalation of beryllium sulfate tetzahydrate at a concentration approximating 90 mg of the salt per cubic meter of air. The animals were exposed 6 hours daily for a two-week period in a small inhalation exposure chamber. *Mortality*, for all species, was 43% of the exposed animals or 20 of 20 mice, 2 of 10 rats, and 2 of 7 hamsters. No deaths occurred among the 2 dogs, 3 rabbits and 14 guinea pigs. *Weight response* data showed that all species save the guinea pig were adversely affected by the exposure. The rat and hamster showed greatest weight depression, losing 11% of their original weight. *Clinical chemical* values indicated renal impairment and some subsequent regeneration in the rabbits. Much less serious kidney damage and hepatic injury in dogs was indicated from biochemical tests performed on both blood and urine. *Hematologic* results showed definite upward trends in the leukocytic count during the second week of exposure. *External symptoms* differed in extent in each species. Thus, ocular opacity developed in the dog, mouse and guinea pig whereas the rat developed râles; other species were unaffected externally. The *histologic* findings showed the effect of the absorption of $BeSO_4 \cdot 4H_2O$ through the respiratory tract of the mouse, rabbit and rat to be pulmonary edema. Hepatic injury was observed in mice dying during the first 9 days, but not thereafter. Renal changes were found in the rabbit and rat with some regeneration of the tubular epithelium in the former species. Marked species variation, but good conformity within each species, was observed in the response to the inhalation of $BeSO_4 \cdot 4H_2O$. The sulfate ion was felt to contribute significantly to the toxicity of the beryllium salt.

The blocking action of magnesium ion on sympathetic ganglia. JOHN B. STANBURY (introduced by OTTO KRAYER). *Department of Pharmacology, Harward Medical School.* If magnesium ion is injected intravenously, or into the arterial blood supply of the ganglia in cats during preganglionic stimulation of the superior cervical ganglion, the stellate ganglion, or the inferior mesenteric ganglion, a brief blockade of impulse transmission occurs. This blockade is located at the ganglion, since the injection has no influence on conduction in the pre- and postganglionic fibers. Magnesium ion has a relaxing action on the nictitating membrane independent of its effect on the superior cervical ganglion, and a decelerating effect on the heart independent of its action on the stellate ganglion. Magnesium ion given intravenously lowers the blood pressure in the intact cat as well as when sympathetic tone has been eliminated by pithing or by giving a constant infusion of epinephrine. This suggests that the blood pressure lowering action of magnesium ion is not wholly attributable to its effect on the sympathetic ganglia.

Potentiation of the effect of antihistaminic agents by iron compounds. GEORGE W. STAVRAKY. *Department of Physiology, Faculty of Medicine, University of Western Ontario, London, Canada.* Noting that Ca^{++} is necessary for blood clotting, neutral salts aid the action of heparin, and iron-containing substances play an important role in intracellular respiration, it was decided to investigate metallic compounds as possible activators of antihistaminic agents. While Ca and K were ineffectual, Ferrous Sulphate, in decerebrated vagotomized cats, increased the effectiveness of small doses of Antistine in counteracting blood pressure changes induced by histamine. Experiments on 8 patients with seasonal hay-fever and asthma due to ragweed showed that administration of Ca or K with antihistaminic drugs was again ineffectual whereas Ferrous Sulphate by mouth in daily doses of 20–45 grains added to Pyribenzamine or Antistine was most effective in alleviating allergic manifestations. Iron also seemed to relieve the lassitude and drowsiness induced by antihistaminic agents; this latter action made it possible to increase the doses of Pyribenzamine and Antistine with further benefit to the patients (50–100 milligrams Pyribenz-

amine sometimes combined with as much as 200 milligrams Antistine, and 0.5–0.6 grams Ferrous Sulphate were given 4–5 times a day without ill effects. As a precaution, iron was given with calcium which is known to reduce the toxicity of iron). One patient, in spite of taking 600 milligrams of Antistine daily, developed asthma which was not materially helped by intramuscular injections of epinephrine in oil. When put on antihistamine-iron therapy he was relieved from asthma within 45 minutes and continued in relative comfort throughout the remainder of the hay-fever season in the face of an increasing pollen count.

Adrenergic blocking properties of certain halogenated ethylamine derivatives. CLEM A. STONE (by invitation), PAUL ACHENBACH (by invitation) and EARL R. LOEW. *Dept. of Pharmacology, Univ. of Illinois College of Medicine, Chicago.* Experiments were made to study the adrenergic blocking action of N-(2-bromoethyl)-N-ethyl-1-naphthalenemethylamine·HBr and β-2-biphenyloxyethyl-butyl-β-chloroethylamine·HCl, which have been shown previously to have the ability to block certain effects of epinephrine as well as histamine (Fed. Proc., **6**: 304, 1947). Adrenergic activity was induced in dogs lightly anesthetized with pentobarbital sodium by (1) occlusion of the common carotids, (2) production of anoxia (N_2 inhalation) and (3) injection of small doses of nicotine; the response in each case was manifested as a rise in blood pressure. The effect of the compounds on the pressor response to injections of epinephrine was also determined. In control experiments made in animals receiving no treatment the above responses remained essentially unaltered for three or four hours. The intravenous doses of N-(2-bromoethyl)-N-ethyl-1-naphthalenemethylamine·HBr and β-2-biphenyloxyethyl-butyl-β-chloroethylamine·HCl were 1 mgm./kgm., and 5 mgm./kgm., respectively. In both atropinized and unatropinized animals both compounds reversed the pressor effects of epinephrine and blocked or reversed the responses to nicotine and anoxia. The pressor responses to carotid occlusion were diminished to a significant degree but were not blocked, possibly because liberation of epinephrine from the adrenal medulla is not concerned in this reaction to the extent that it is in responses to nicotine and anoxia. This evidence, in conjunction with results of previous studies, indicates that N-(2-bromoethyl)-N-ethyl-1-naphthalenemethylamine·HBr is a more potent adrenergic blocking compound than β-2-biphenyloxyethyl-butyl-β-chloroethylamine·HCl and that the former compound is also highly effective in antagonizing histamine.

Effects of alterations in body temperature on properties of convulsive seizures in rats. EWART A. SWINYARD (by invitation) and JAMES E. P. TOMAN. *Departments of Pharmacology, Pharmacy, and Physiology, University of Utah, Salt Lake City, Utah.* Threshold and pattern of electrically and chemically induced convulsions were studied in rats whose body temperature was experimentally varied by environmental heating or cooling. *Seizure threshold.* Electroshock threshold (60 cycle A.C., 0.2 sec. duration) for minimal seizures was decreased by lowering and increased by elevating body temperature. The Q_{10} was 1.5 at 25°–35°C. and 1.8 at 35°–45°C. The results suggest an absolute increase in electrical excitability at reduced temperatures, modified by an increase in time constant of excitation. The thresholds for Metrazol and Picrotoxin seizures were also reduced by lowering body temperature (Q_{10} approximately 2). Extremes in body temperature (below 25°C. and above 43°C.) were convulsant to some animals. *Recovery.* The effect of body temperature on recovery rate of minimal seizure threshold following maximal seizures was studied. The time for recovery to 150% of threshold was approximately doubled by a 10°C. reduction in body temperature (Q_{10} approximately 2). *Seizure Pattern.* The $\log_{10}$ of maximal seizure duration varied linearly with the reciprocal of absolute temperature, giving a Q_{10} of 2.8. The tonic extensor component was decreased in relative duration either by lowering or elevating body temperature, and was abolished below 27° and above 42°C.

Some metabolic products of para-aminobenzoic acid. CELIA WHITE TABOR (by invitation), JUDITH BAILY (by invitation) and PAUL K. SMITH. *From the Department of Pharmacology, The George Washington University School of Medicine, Washington, D. C.* In continuation of our studies on the fate of para-aminobenzoic acid, glycuronate excretion was determined following the oral administration of para-aminobenzoic acid or the sodium salt, using the carbazole method recently described by Dische (J. Biol. Chem. **167**, 189–198 (1947)). In patients given single six gram doses of the drug or smaller doses every two hours, relatively large quantities (up to 2.7 gm.) of apparent glycuronate were present in twenty-four hour urine specimens. The changes in the excretion rate of this substance were roughly parallel to the change in the excretion rate of free amine. Using the countercurrent distribution method of Craig (J. Biol. Chem. **155**, 519–34 (1944)) to supplement other methods, it was determined that the metabolic products of para-aminobenzoic acid probably include the free acid, para-aminohippuric acid, the acetylated forms of each, and a glycuronate derivative of a free amine. These studies suggest that the metabolism of this drug is similar to that of benzoic acid (Snapper, Am. J. Med., **II**, 327–333 (1947)) and some of the sulfonamides.

Urinary excretion of histamine following oral administration, employing an improved colorimetric method. HERBERT TABOR and SANFORD M.

ROSENTHAL. *Division of Physiology, National Institute of Health, Bethesda, Maryland.* A colorimetric method for histamine, based upon a diazo reaction with 4-nitroaniline, has been developed. By the use of a solvent (methyl isobutyl ketone) the color is extracted and then brought to a pH of 7.7 by shaking with barbital buffer. This procedure is sensitive to approximately 0.5 gamma of histamine base, and results upon animal tissues indicate a satisfactory degree of specificity. Anrep has shown that histamine can occur in the urine in free and conjugated form. A corresponding study of urinary histamine following oral administration was carried out in several species of animals. Up to 35 per cent of the amount given could be recovered. There was considerable species variation in the total excretion, and in the amount conjugated.

Some effects of ouabain and/or calcium in mice. ROBERT TARAIL and W. LANE WILLIAMS (Introduced by R. N. BIETER). *Departments of Anatomy and Medicine, University of Minnesota.* Mice (av. wt. 25 gms.) were daily injected subcutaneously or intraperitoneally with (doses expressed in mg. per 100 gms. body weight): (a) ouabain, 0.5 to 2 mg., for 4 to 36 days; or (b) calcium chloride, 33 to 50 mg., or calcium gluconate, 100 to 200 mg., for 13 to 19 days; or (c) with combinations of ouabain and calcium in amounts representative of the range of doses listed in "a" and "b." Total daily amounts of ouabain greater than 0.25 mg. were injected in divided doses over a period of 1 hour since larger single doses often resulted in immediate death. Most of the treatments with one of the compounds and all of the combined treatments produced small focal areas of myocardial necrosis. Mice receiving ouabain (alone) showed in addition to myocardial lesions some edema, venous and capillary congestion in kidneys, liver, and lungs. In addition to these changes treatment with calcium or combinations of calcium and ouabain produced necrosis and hemorrhage in the livers and kidneys. An increased dual toxicity of combinations of ouabain and calcium was suggested by reduction in life expectancy in relation to that observed in mice on single treatment with either calcium or ouabain. However, any increase in the toxicity and lethality of combinations of ouabain and calcium could not be quantitatively correlated with increased total tissue damage (heart, liver, kidneys, and lungs) and such doses demonstrated no clear synergistic or cumulative effects in producing morphological myocardial injury.

The pharmacology of organic thiocyanates. SALAH A. ABDEL TAWAB (introduced by JOHN C. KRANTZ, JR.). *Department of Pharmacology, Univ. of Maryland, School of Medicine, Baltimore.* The study of some organic thiocyanates revealed the depressor action of these compounds especially with p-thiocyanophenol. Further study of this substance showed that it diminishes the oxygen consumption in white albino rats together with pathological changes in the thyroid gland analogous to those of colloid goiter. The insolubility in water and the high toxicity of this compound has led us to suggest the synthesis of thiocyanobenzoic acid derivatives, the sodium salts of which are soluble in water and the benzoic derivative is comparatively non-toxic. Sodium p-sulfocyanobenzoate is found to have a prolonged depressor action with slow return to normal and marked respiratory stimulation—an action which is believed to be central in origin. Comparative study with sodium thiocyanate showed that the sulfocyanate radical is about 80 times more potent than the radical in polar combination with an alkali cation.

Factors affecting the use of lung thromboplastin in the determination of prothrombin time. BERNARD G. H. THOMAS (by invitation), JACQUELINE SPEARS (by invitation), and CHARLES R. LINEGAR. *Pharmacological Development Division, E. R. Squibb & Sons, New Brunswick, New Jersey.* Dried thromboplastin from rabbit lung was found to be more stable at room temperature and more potent, and to give better correlation of clotting time with various prothrombin levels in undiluted and saline diluted plasma than thromboplastin from rabbit brain. Lung thromboplastin showed greater activity when extracted with an aqueous solution containing both sodium and calcium chloride rather than with one containing only sodium chloride. The addition of 0.4 per cent phenol to the extracting fluid increased in most cases the activity of the extract, and preserved the thromboplastic activity for approximately 24 hours when the extract was stored at room temperature. Factors lengthening the prothrombin time were found to be: the removal of part of the suspended thromboplastin by centrifugation of the extract, the addition of inadequate or excessive amounts of calcium chloride in the test, the use of more than the required amount of oxalate in the collection of blood samples, the presence of clots in the blood sample to be tested, and undue delay in performance of the test. In order to evaluate prothrombin times properly, the tests should be made with a standardized thromboplastin under carefully controlled conditions.

Effect of adrenolytic agents on the response to pressor substances in the domestic fowl. RONALD M. THOMPSON (by invitation) and JULIUS M. COON. *Dept. of Pharmacology, University of Chicago.* It has been shown that dibenamine-HCl causes a reversal of the pressor response to epinephrine in mammals. Intravenous dose of 20 mg./kg. of dibenamine-HCl neither reversed nor depressed the pressor responses to small or large doses of epinephrine in the domestic fowl. Dibenamine also did not affect appreciably the pressor responses (in amounts equivalent in pressor effect to 0.1 cc. of

epinephrine, 1:100,000) to benzedrine, cobefrin, ephedrine, neosynephrine, synephrine, tyramine or nicotine in the fowl. However, dibenamine-HCl markedly diminished the pressor response to the equivalent amount of Varon (2,4-dimethoxy-alpha-phenyl-beta-methylamino ethanol. Priscol and dihydroergotamine, unlike dibenamine-HCl, diminished (though did not reverse) the pressor response to epinephrine. This is interpreted as indicating a difference in the mechanism of action of these two adrenolytic drugs from that of dibenamine-HCl. The domestic fowl was markedly resistant to the toxic effects of epinephrine. As much as 30 mg./kg. of epinephrine (1:100) injected rapidly intravenously did not cause the immediate death that occurs in mammals given much smaller amounts. However, doses upward from 2.5 mg./kg. of epinephrine caused immediate prostration, followed in about 10 minutes by extreme difficulty in the expiratory phase of respiration, and death in 10 to 120 minutes. Death appeared to be the result of increasing respiratory difficulties. An intravenous dose of 10 mg./kg. of epinephrine one hour after 20 mg./kg. of intravenous dibenamine-HCl caused no prostration, only slight respiratory difficulty, and full recovery in the majority of chickens. Thus, dibenamine-HCl protected the domestic fowl against the toxic effects of large doses of epinephrine although it did not affect the pressor response to epinephrine.

Effect of convulsant and anticonvulsant agents on acetylcholine metabolism and effector organs' acetylcholine sensitivity. CLARA TORDA and HAROLD G. WOLFF. *New York Hospital and the Departments of Medicine (Neurology) and Psychiatry, Cornell University Medical College, New York, N. Y.* It has been reported that acetylcholine applied locally to the motor areas of the cerebral cortex or administered parenterally in animals induces changes in the electroencephalogram resembling those observed during 'grand mal' attacks in man. In the following it was ascertained whether or not the convulsion-inducing agents (pentamethylene tetrazol, picrotoxin, strychnine, morphine, camphor, scilliroside, digitoxin) (a) increase the synthesis of acetylcholine (increased activity of choline acetylase); (b) decrease the hydrolysis of acetylcholine (decreased activity of cholinesterase); and (c) increase the sensitivity of effector organs to acetylcholine. All of these measures, together or separately, may induce a temporary accumulation of acetylcholine. It was also ascertained whether or not the anticonvulsant agents (hydantoin, methyl-phenyl-ethyl hydantoin, diphenylhydantoinate sodium, trimethadione, sodium bromide, phenobarbital, barbital sodium, pentobarbital sodium, diallylmalonylurea, iso-amyl-ethyl-barbiturate sodium) (a) decrease acetylcholine synthesis; (b) increase the hydrolysis of acetylcholine; and (c) decrease the sensitivity of effector cells to acetylcholine. These measures, together or separately, may induce a temporary decrease of acetylcholine. The results show that most of the convulsion-inducing agents cause an accumulation of acetylcholine and most of the anticonvulsant agents cause a decrease in the concentration of acetylcholine. There are, however, convulsion-inducing and anticonvulsant agents that do not modify acetylcholine metabolism in the above demonstrated manner. It seems that accumulation of acetylcholine per se is not the primary factor in the induction of convulsive seizures nor is lack of acetylcholine the primary factor in prevention of convulsive seizures.

The quantitative estimation of theophylline in blood. EDWARD B. TRUITT, JR. (introduced by JOHN C. KRANTZ, JR.). *Department of Pharmacology, Univ. of Maryland, School of Medicine, Baltimore.* A colorimetric method for the quantitative estimation of theophylline in blood has been developed. The method is based on the formation of a red color through the coupling of a diazo compound, the diazotized 5-amino-2 benzoylamino-1,4 diethoxybenzene, with an alkaline oxidation product of theophylline. The reaction produces a comparatively stable red color that will detect theophylline in blood levels of 0.1 mgm.% or more. The method has been applied to patients undergoing theophylline therapy.

The spreading of Evan's blue injected intradermally. RALPH UBER (by invitation), PATRICIA COLLINS COWDERY (by invitation) and GEORGE E. FARRAR, JR. *Dept. of Medicine, Temple Univ. School of Medicine, Philadelphia, Pa.* Following the intradermal injection of 0.1 cc. of 0.24% Evan's blue in aqueous 0.9% sodium chloride solution, the longest and shortest diameters of the blue area were measured. The spreading index was calculated,—square of the median radius at 3 and 24 hours divided by this figure for the initial wheal. The degree of spreading varied greatly among both healthy and diseased persons but was repeatedly similar in an individual. The average spreading index in a group of 30 healthy, young adults (15 men, 15 women) was 4.5 at 3 hours, with a range of 1.5 to 10.7, and 10 at 24 hours, 4.5 to 23.8. The degree of spreading at 3 hours seemed to be little affected by factors such as exercise and weather which may alter the reading at 24 hours considerably. Patients with rheumatic fever showed no gross differences; 5 cases in the active phase had an average index of 3.4 at 3 hours; 5 inactive cases averaged 4.5. Fifteen cases of active rheumatoid arthritis had an average index of 11.2 at 24 hours with a range of 5.4 to 29.6. Ten patients with far advanced pulmonary tuberculosis had an average of 6.6 and 10 minimal cases 5.1 at 3 hours. Pregnant patients before and after delivery and cases of periarteritis nodosa, disseminated lupus

erythematosis and osteoarthritis have been studied. No correlation has been found between spreading index, erythrocyte sedimentation rate and the inhibiting effect of the blood serum on the action of hyaluronidase derived from hemolytic streptococci of group A.

The estimation of alpha and beta estradiols and of estrone and equilin in binary mixtures. ERNEST J. UMBERGER and JACK M. CURTIS (introduced by ARNOLD J. LEHMAN). *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington 25, D. C.* The behavior of several natural estrogens toward various concentrations of sulfuric acid has been studied. These studies have provided a method for the identification and quantitative estimation of the estrogens after their partial separation by chromatography. Beta-estradiol may be determined by heating with a solution of 30% sulfuric acid and 20% n-butyl alcohol in water on a boiling water bath for 6 minutes and reading the color at 524 mmu. Total estradiols are determined by heating with 90% sulfuric acid for 15 minutes and reading at 444 mmu. Alpha-estradiol is determined by difference. Estrone and equilin may be determined simultaneously by heating with 90% sulfuric acid for 5 minutes and reading at 450 and 480 mmu. The proportion of estrone or equilin can then be calculated by simultaneous equations or by applying the formula of Knudson, Meloche, and Juday (An. Ed., Ind. Eng. Chem. **12**, 715, 1940).

Efficacy of atropine in children. K. R. UNNA, K. GLASER (by invitation), E. LIPTON (by invitation), and P. PATTERSON (by invitation). *Departments of Pharmacology and Pediatrics, University of Illinois College of Medicine, Chicago 12, Illinois.* Effects of graded doses of atropine following oral and subcutaneous administration were determined in children aged 1 month to 12 years. The suppression of the stimulation of salivary flow by chewing gum over a specified 5 minute period was chosen as the criterion for threshold effects in older children. The threshold doses determined by the chewing test were identical with those suppressing the flow of saliva elicited by small doses of methacholine injected subcutaneously. In children below the age of 3 years, minimum effective doses of atropine were estimated by the saliva flow after methacholine. Although appreciable individual variations, especially on oral administration, were sometimes encountered, the average threshold doses in the various age groups (1–12 months; 1–3 years; 3–6 years; and 6–12 years) were found to lie in a narrow range from 15 to 23 micrograms after oral doses and from 4 to 6 micrograms after subcutaneous doses of atropine per kgm. body weight. Slightly larger doses often caused flushing and febrile reactions in small children. Still larger doses were required to raise the pulse rate and to dilate the pupils. The implication of these studies on the concept of physiological "vagotonia" in young children will be discussed.

A comparison of curare alkaloids. E. F. VAN MAANEN (introduced by OTTO KRAYER). *Department of Pharmacology, Harvard Medical School.* The preparations studied included d-tubocurarine, c-toxiferine II and an extract of the bark of Strychnos Toxifera. These preparations decrease the sensitivity of the isolated rectus abdominis of the frog to acetylcholine. The shape of the dose-response curve suggests a competitive reaction between acetylcholine and the curare alkaloids. The relative activity of the alkaloids on the rectus abdominis does not correspond to their relative lethal dose in mice, injected intravenously. Of the three alkaloids named the relative activity in the rectus was, respectively, 1, 2.5 and 0.3. The corresponding LD 50's in mice were 1, 0.62, and 0.044 (for the extract, the dry weight was used in calculating these results; 1 gram of extract, dry weight, corresponds to 3 grams of bark). In rats anesthetized with urethane, twitches from the Achilles tendon on maximal stimulation of the sciatic nerve were studied after intravenous injections of the alkaloids. From the time course and extent of the neuromuscular block deductions can be made concerning rates of binding of the alkaloids to muscles and rates of elimination of the alkaloids from the body.

The rabbit "head-drop" method for the biological assay of curare and its alkaloids. ROGER F. VARNEY (by invitation), CHARLES R. LINEGAR and HORACE A. HOLADAY (by invitation). *Biological and Chemical Laboratories, E. R. Squibb & Sons, New Brunswick, New Jersey.* In the early development of standardized curare, the classical methods of assay failed to serve as a reliable index of potency of various curare preparations in humans. In this respect the rabbit "head-drop" method which employs muscular relaxation in an intact mammal as the criterion of curare activity has proven best. To reach "head-drop" a rabbit, tied belly down, is given an interrupted intravenous injection of a solution containing 2 units per ml. ± 10 per cent *via* a microburette at the rate of 0.1 ml. every 15 seconds until muscles supporting the head becomes sufficiently relaxed to prevent its being raised when the back is stimulated. The end point is sharp and reproducible. The "head-drop" dose in different rabbits even of the same weight varies widely, therefore it is of utmost importance to compare the potency of an unknown curare with that of a standard in the same animal. Since, following preliminary curarizations, the daily "head-drop" dose requirement of each rabbit is quite constant, the assay is applied as a two day "cross-over" test. In 550 determinations of the ratio of activity of standard *versus* unknown in randomly selected rabbits,

the standard deviation of a single ratio was calculated and on this basis the standard error for the mean of ten ratios was 3.9 per cent. The curare unit originally defined as the activity of 1 mg. of a desiccated curare powder was later shown to be equivalent to about 0.15 mg. of the "pentahydrate" of *d*-tubocurarine chloride.

Comparative studies of the toxicity and physiological action of chlorinated methanes with reference to their physical and chemical characteristics. W. F. VON OETTINGEN, C. C. POWELL (by invitation), N. E. SHARPLESS (by invitation) and L. J. PECORA (by invitation). *From the Laboratory of Physical Biology, National Institute of Health, Bethesda, Md.* The LD_{50} for mice by inhalation for 7 hours of mono-, di-, tri-, and tetrachloromethane was determined, and the effect of these chlorinated methanes on the circulatory and respiratory apparatus, the nervous system, and their concentration in heart, liver, brain, and blood with exposure to concentrations of 15000 p.p.m. was studied in dogs. The reaction rate constant and the partition coefficient in a system oleic alcohol/water was determined. It was shown that monochloromethane is readily decomposed and that this is also reflected in its physiological action and distribution in the organs studied. Di-, tri- and tetrachloromethane lowers increasingly the blood pressure in the order given, only the two latter cause direct cardiac depression under the conditions studied. Whereas monochloromethane causes first a marked stimulation of the respiration and only a depression after a latent period of several hours, di-, tri-, and tetrachloromethane cause a depression, increasing in the order given. Whereas the depressant effect of monochloromethane on the nervous system is very moderate, this increases intensity from di-, over tetra-, to trichloromethane. The organ distribution of the chlorinated methanes shown that in the case of tetrachloromethane there is a distinct and with trichloromethane a less marked accumulation in the central nervous system. The parallelism between the physiological behavior and certain physical-chemical characteristics is discussed.

The paralytic and lethal action of myanesin, pentobarbital and combinations of these agents in mice. H. A. WALKER (by invitation) ARTHUR P. RICHARDSON, P. LOEB (by invitation) and J. PEROG (by invitation). *Department of Pharmacology, Emory University School of Medicine, Emory University, Georgia and Division of Pharmacology, Squibb Institute for Medical Research, New Brunswick, New Jersey.* We have been unable to confirm the conclusions reached by Berger and Bradley (Brit. J. Pharmacol. **1**, 265 (1946)) that the combined depressant effect of myanesin and a barbiturate is greater than might be expected from the individual activities of each agent. Loss of righting reflexes was taken as a criterion of paralysis. White mice of 18–22 grams were injected intraperitoneally with varying doses of myanesin and sodium pentobarbital, and various combinations of these two agents. Any animal unable to right himself within 15 seconds after being placed on his back was considered paralysed. All animals were held for 24 hours and deaths noted for this period of time. The PD_{50} for myanesin alone was found to be 182 mgms. per kilo, and for sodium pentobarbital 43 mgms. per kilo. In mice injected with 90 mgms. per kilo of myanesin (50% of PD_{50}), 19.5 mgms. per kilo of pentobarbital (45.5% of PD_{50}) were required to produce paralysis. This result is within 5% of the expected additive effect. In other series of mice receiving from 12.5 to 75% of the PD_{50} of myanesin plus the barbiturate similar results were obtained. The lethal effects of combinations of these two agents were also found to be purely additive.

Comparative increase in ventricular contractile force produced by several cardiac glycosides. R. P. WALTON, M. F. PATTON (by invitation), H. P. JONES (by invitation) and J. S. LEARY (by invitation). *Dept. of Pharmacology, Medical College of South Carolina, Charleston.* Five cardiac glycosides and tincture of digitalis were compared at dose levels ranging from 0.25 to 1.3 cat units per kgm. Determinations of ventricular contractile force (isometric systolic tension of a section of the right ventricle in open-chest dog preparations) were made according to the method described in J.P.E.T., 89: 26, 1947. The glycosides used were ouabain, thevetin, scillaren, strophanthidin-acetate and lanatoside C (Cedilanid). Total doses were administered during a period of 30 minutes at uniform rates of infusion. With doses of 0.25 cat units per kgm., maximum effects were obtained in an average of 42 minutes from the beginning of the infusion. Increasing doses progressively produced shorter intervals before development of maximal effects. With doses of 1.3 cat units per kgm., maximum effects were obtained in an average of 22 minutes. Effects developed most rapidly with thevetin and most slowly with tincture of digitalis and with lanatoside C. Comparison of the increases in contractile force produced by the individual members of this series supports the conception of their basic similarity. Possible deviations, which are being further examined, indicate, in the lower dosage range, a greater than average effect with thevetin and a less than average effect with tincture of digitalis. Based on data obtained from 15 to 20 experiments with each glycoside, the average maximal increases in contractile force (expressed as percentage increase over that of the control period) were as follows: 0.25 cat units, 31%; 0.50 cat units, 46%; 0.75 cat units, 60%; 1.0 cat units, 93%; 1.3 cat units, 100%.

Hypnotic properties and toxicity of 2-ethyl-3-propylglycidamide. MARSHALL R. WARREN (by

invitation), CHARLES R. THOMPSON (by invitation), and HAROLD W. WERNER. *Pharmacology Dept., Research Labs., The Wm. S. Merrell Co., Cincinnati, O.* 2-Ethyl-3-propylglycidamide has a central depressant action in experimental animals generally resembling that of shortacting barbiturates grossly, and it also antagonizes metrazol-induced convulsions. Intravenous studies in rabbits demonstrate that the acute LD_{50}s for 2-ethyl-3-propylglycidamide, pentobarbituric acid, and ethylisoamylbarbituric acid are > 360, 34, and 55 mgm./kgm. respectively; and minimal hypnotic doses (smallest doses which caused 50 per cent or somewhat more of the animals receiving them to be on their sides with heads down) are 70, 10, and 13 mgm./kgm. All three compounds have similar durations of action since the intravenous administration of two minimal hypnotic doses causes an average narcosis of 1.4 to 1.6 hours. Oral studies in rats show that the acute LD_{50}s for 2-ethyl-3-propylglycidamide, pentobarbituric acid, and x-monobromoisovalerylurea (Bromural) are 1.25, 0.125, and 1.0 gm./kgm. respectively; and minimal hypnotic doses are 0.50, 0.063, and 0.50 gm./kgm. All three substances are similar in duration of action following oral administration in rats since one minimal hypnotic dose produces a median narcosis of 3.3 to 4.5 hours. Repeated daily administration of 2-ethyl-3-propylglycidamide, in doses equal to one-tenth and two-fifths of the oral minimal hypnotic dose for rats, to rats, dogs, and monkeys for periods of four to eight weeks gave no evidence of toxicity or cumulative action. Hematologic studies on the experimental animals revealed no alteration of the blood picture from that of the control animals. Microscopic examination of tissue sections showed no pathologic changes which could be attributed to drug administration.

Effect of some central nervous system stimulants and depressants on the activity of succinic dehydrogenase. D. T. WATTS (introduced by C. L. GEMMILL). *Department of Pharmacology, University of Virginia, Medical School, Charlottesville, Virginia.* The effect of selected central nervous system drugs on succinic dehydrogenase activity in vitro has been determined, using the Thunberg technique. The rate of methylene blue reduction by the enzyme preparation from washed frog muscle with sodium succinate as substrate was followed with a photoelectric colorimeter or by determining an end point visually. Barbital, pentobarbital sodium, ethyl urethane, chloral hydrate and sodium bromide gave no inhibition at final concentrations of 1.2×10^{-3} to 4.0×10^{-2} M. Caffeine produced inhibition at concentrations as low as 1.3×10^{-3} M. and gave complete inhibition at 2.1×10^{-2} M. Similar inhibition was observed with theophylline. A technique has been developed for passing volatile anesthetics into evacuated Thunberg tubes either in the gaseous state or by allowing the liquid to distill into the tube. Chloroform inhibits this dehydrogenase. This loss of activity can be partially restored by boiling off the chloroform by additional evacuation.

The fate of p-aminosalicylic acid in the animal body. E. LEONG WAY, ROWENA WEISS (by invitation), DONALD L. HOWIE (by invitation) and PAUL K. SMITH. *From the Department of Pharmacology, The George Washington University School of Medicine, Washington, D. C.* The absorption, distribution and excretion of p-aminosalicylic acid (PASA) was investigated in the rat, dog and man by determining free amine, conjugated amine and free phenol. There is little or no storage of PASA in rats after intravenous or oral dosage (200 mgm. per kgm.). Within four hours, the compound is found chiefly in the urine, with a small fraction in the gastrointestinal tract. By far the highest tissue concentrations are attained in the kidney, then in the lung and liver, with the latter yielding appreciable values for conjugated amine. In dogs, about ninety per cent of an intravenous 100 mgm. per kgm. dose of PASA can be recovered in the urine as free amine or free phenol after eight hours. The rate of fall or plasma levels closely parallels the rate of appearance in the urine. In humans, after an oral four gram dose, plasma levels rise rapidly to a maximum of approximately 10 mgm. per cent and then fall rapidly, with over ninety per cent of the total dosage being recovered as total amine or free phenol. Even after five or six single 2.5 gram doses at six hour intervals, over eighty per cent of the total amount given can be accounted for within six hours after the last dose, with about fifty to sixty per cent being present as conjugated amine. On comparing the partition coefficients of urinary products and of added PASA in ether and acetate buffer (pH 3.4), it was found that the free amine in urine is present in a more water-soluble form.

Metabolism of rat heart slices. J. LEYDEN WEBB (by invitation), PAUL R. SAUNDERS (by invitation) and CLINTON H. THIENES. *Department of Pharmacology and Toxicology, School of Medicine, University of Southern California, Los Angeles, California.* The respiration of slices of rat ventricular myocardium was studied in the presence of various substrates and inhibitors. Substrates fall roughly into four classes according to the ability of the heart to utilize them: increase respiration over 200 per cent (succinate), from 50–100 per cent (citrate, oxaloacetate), from 10–30 per cent (pyruvate, lactate, malate, hydroxybutyrate) and no effect (ketoglutarate, glucose, glucose-1-phosphate, hexose-diphosphate). Glucose has no immediate effect but after two to three hours the respiration begins to rise until it is higher than initially, thus glucose maintains respiration but requires an appreciable time to be utilized. Malonate, at a con-

centration (0.02M) completely inhibiting succinate oxidation, inhibits the endogenous respiration of heart slices 30 per cent. With concentrations of malonate between 0.002M and 0.02M the respiration rises from its originally depressed value for 90 minutes and then falls off more slowly than control; at 90 minutes the respiration may be higher than control. A type of respiration that is malonate-insensitive seems to increase during this period: preliminary evidence indicates the malonate is responsible for this. This malonate "hump" occurs only in the presence of calcium. Fluoride inhibition of cardiac respiration increases with fluoride concentration up to 0.01M but then decreases to a minimum when fluoride is 0.05M and then increases again as fluoride concentration raised. Iodoacetate and phlorizin are very potent inhibitors of cardiac respiration while pyrophosphate is a relatively week inhibitor.

The influence of N-(2-bromoethyl)-N-ethyl-1-naphthalene-methylamine on the vasopressor response of a series of amines. J. A. WELLS and DAVID P. RALL (by invitation). (*From the Department of Pharmacology, Northwestern University Medical School, Chicago, Illinois*). Reversal of the pressor action of epinephrine characteristically follows the administration of potent sympatholytic drugs. Occasionally this same phenomenon has been observed with other pressor amines, but some difference of opinion exists as to which compounds are reversed and some are claimed never to be reversed. The compound, N-(2-bromoethyl)-N-ethyl-1-naphthalene-methylamine (SY 28), recently synthesized and supplied to us by Doctors Rieveschl, Fleming and Coleman, has been shown by Dr. Earl Loew to be a potent sympatholytic drug. We have been interested in knowing which of the pressor amines are blocked or reversed by this drug. Following the slow intravenous administration of 1-5 mg./kg. of this compound to nembutal-anesthetized dogs, cobefrine, adrenalone and tyramine have shown marked reversals similar to epinephrine. Ephedrine, neosynephrin, amphetamine, privine and arterenol have also been reversals similar to epinephrine. Ephedrine, neosynephrine, amphetamine, privine and arterenol have also been reversed. Propadrine, paredrine and paredrinol have been reversed but only diminution or blocking of the pressor response is usually obtained with these compounds. The pressor response to pitressin and the depressor response to isopropyl epinephrine are unaffected by the sympatholytic agent. With increasing doses of the amines, decreasing pressor responses precede reversal.

Actions of podophyllum derivatives. D. G. WENZEL (by invitation), O. S. ORTH, R. T. CAPPS (by invitation), and A. H. UHL (by invitation). *Depts. of Pharmacy and Pharmacology, University of Wisconsin, Madison.* Recently the resin of Podophyllum, a cathartic obtained from *Podophyllum peltatum Linné*, has received attention for its effects on body cells, particularly in mitotic division, and on venereal warts. From the resin various compounds may be isolated by the use of solvents. Certain of these compounds, including a yellow Quercetin pigment and picropodophyllin, have been tested on mice, rats, rabbits and dogs for acute and chronic toxicities, and effects on the respiratory, cardiovascular and hemopoietic systems. Difficulties have been encountered in obtaining suitable solutions or suspensions with small enough particle size to permit intravenous administration of all the fractions. Toxicities based on oral, intraperitoneal or intravenous routes of administration will be presented for certain of the species. It appears that for the Quercetin compound, initial toxic effects are due to actions on the respiratory system, with makred circulatory depression soon following. Dosages repeated at intervals of ten or fifteen minutes show evidence of tachyphylaxis. Clinical actions of the individual fractions of *Podophyllum peltatum L.* will be reported.

Studies on diffusion respiration VI. Changes in kidney function in dogs. R. W. WHITEHEAD, D. L. G. BESHORE (by invitation), W. B. DRAPER, JOSEPH N. SPENCER (by invitation) and THOMAS M. PARRY (by invitation). *From the Department of Physiology and Pharmacology University of Colorado Medical Center.* Preliminary observations on ten dogs in diffusion respiration under pentothal (thiopental) in which urinary output was recorded, revealed that anuria developed in all animals during respiratory arrest. In an additional series of ten dogs under thiopental anesthesia, alterations in kidney function were studied. Urinary output, chloride excretion, urinary and femoral venous blood pH and arterial blood pressure were recorded before, for fifty minutes during, and following prolonged respiratory arrest. Five of the animals were not given additional fluids; the other five were given an infusion of normal saline intravenously throughout the experiment. *Results:* Anuria developed in all dogs during respiratory arrest. Often this occurred promptly with the cessation of respiration. There was no appreciable difference in the time of onset of anuria in the hydrated and non-hydrated groups. In the non-hydrated group the urinary chlorides rose from an average control value of 187 mg. per cent ot 517 mg. per cent two hours after respiration was resumed. In the hydrated group the chlorides fell from a control of 3090 mg. per cent to about 25 mg. per cent. Urine flow returned within an average period of 72 seconds after respiration was resumed. Arterial blood pressure values were sufficient to provide an effective glomerular filtration pressure throughout the experiments. Femoral venous blood pH fell from

an average control value of 7.33 to 6.67 at forty-five minutes of respiratory arrest. The average pH at the end of 1½ hours post-diffusion was 7.34. Urinary pH changes paralleled those of the blood during the pre- and post-diffusion periods.

The effect of "denervation" on the response of intestinal fistulae to several drugs. R. M. WHITROCK (by invitation), H. L. TIECHE (by invitation), and M. H. SEEVERS. *Department of Pharmacology, University of Michigan.* Observations were made on the response of serial innervated and "denervated" Thiry-Vella jejunal fistulae in normal, unanesthetized dogs. The finding of Herrin and Meek that distention of innervated loops at 90 mm. Hg pressure reflexly induces vomiting and other signs of intestinal obstruction was confirmed. The "denervated" loop is more sensitive (3–4 times) than the innervated loop to the inhibitory action of epinephrine, as noted previously by Youmans. We have found a quantitatively similar sensitivity in the "denervated" loop to the inhibitory action of atropine and tetraethylammonium. If it is assumed that the normal functional activity of the intestine is a resultant of the opposing excitatory and inhibitory actions of the two autonomic divisions; and that the intestine is constantly influenced by a circulating epinephrine-like substance; then the observations on atropine and tetraethylammonium indicate that, whereas socalled "denervation" of intestinal loops by section of all nerve fibers at the vascular pedicle results in complete isolation of the structure from all central nervous connections such a procedure does not effect real denervation but only results in (1) complete loss of sympathetic innervation (after degeneration of the postganglionic fibers). (2) Section and degeneration of the preganglionic fibers of the parasympathetic division, the ganglion and postganglionic fibers remaining intact. (3) Sensitization of sympathetic effector cells and probably sensitization of parasympathetic ganglia as noted by Rosenbleuth and Cannon for the nictitating membrane.

Reactions of chronic decorticated dogs during a cycle of addiction to methadon. ABRAHAM WIKLER. *Research Department, U. S. Public Health Service Hospital, Lexington, Kentucky.* In 2 chronic decorticated dogs subcutaneous injection of 2.0 to 5.0 mg./kg. of methadon ("amidone", "10820") regularly reduced spontaneous activity, depressed sham rage responses to pinching or restraint, elevated tooth pain reaction threshold, lowered body temperature and cardiac rate. In one such preparation, 43 days after completion of decortication, methadon was injected subcutaneously every 6 hours for 60 days, the dose being increased rapidly from 2.0 to 5.0 mg./kg. A high degree of tolerance developed to the analgetic, sham rage depressant and temperature lowering effects, but tolerance to the depressant effects on spontaneous activity was less marked. As addiction progressed pre-injection restlessness and hyperirritability were observed. Two attempts at abrupt withdrawal were abandoned because the preparation exhibited extreme restlessness, marked loss of irritability, profuse salivation, vomiting, tachycardia, fever, weak pulse and gasping respirations. After a 2-day withdrawal, restlessness, hypoirritability, persistent rooting and gnawing at the floor of the circular cage were exhibited for about two weeks after which there was a gradual return to the preaddiction state. In another preparation, 6 months after completion of decortication, methadon 2.0 mg./kg. was substituted for morphine after tolerance to the latter had been established. This dose was injected subcutaneously every 6 hours for 34 days. Partial cross tolerance to morphine was observed. On abrupt withdrawal of methadon, an abstinence syndrome was observed which was milder but qualitatively the same as that described above. The methadon abstinence syndrome in both preparations was indistinguishable from that of morphine.

Effects of electroshock convulsions on chronic decorticated cats. ABRAHAM WIKLER and KARL FRANK (by invitation). *Research Department, U. S. Public Health Service Hospital, Lexington, Kentucky.* In chronic decorticated cats convulsions were induced by passage of 200–450 milliamperes of 60 cycle A.C. from vertex to palate. Immediately after each seizure and for ½ to 4 hours afterward sham rage responses (chiefly facio-vocal) to pressure-pain stimuli applied to the tail were markedly reduced while other sham rage responses (springing, clawing, lashing of tail) evoked by non-nociceptive stimuli were unaffected or enhanced. In 3 of the preparations licking responses to tactile stimulation of the perineal region were temporarily depressed after each seizure. Righting reflexes returned within a few minutes after each convulsion while pulse rate and rectal temperatures were not altered significantly. Apnea occurred during the seizures and was followed by transient hyperpnea. No cumulative or new effects were observed after daily electroshock convulsions for 5 to 9 days. In terminal experiments the preparations were curarized and maintained on artificial respiration. Electrical activity was recorded from vertex and sphenoid screw leads immediately after electroshock induced as above. The electroencephalographic patterns were characterized by bursts of 6–21 per second rhythms interrupted by silent intervals finally terminating after a more prolonged 15–18 per second rhythm. In places fast and slow wave sequences were seen. This was more marked in preparations which were morphinized prior to electroshock. Neuropathologic studies of the remaining brains revealed no residual neocortex.

The microscopic changes did not differ in degree or kind from those seen in other chronic decorticated cats which were not subjected to electroshock convulsions.

Subtilin in blood after parenteral administration. ROBERT H. WILSON, J. C. LEWIS and E. M. HUMPHREYS (by invitation). *From the Pharmacology Division and the Western Regional Research Laboratory of the Bureau of Agriculture and Industrial Chemistry, U. S. D. A., Albany 6, California.* Subtilin, subcutaneously, has very low toxicity, 3 gm./kg. of purified subtilin does not kill mice (Anderson and Chin, Wilson, unpublished), due to the physical characteristics of the substance. In physiological saline or serum its solubility does not exceed 0.05–0.06 gm./100 ml. Subcutaneously administered subtilin precipitates at the site of injection. Intramuscularly administered subtilin likewise is precipitated and absorbed slowly. Amounts up to 100 mg./kg. have been injected into the thigh muscles of rabbits and cup plate assay of periodic blood samples has indicated no more than 2 ppm. of subtilin in whole blood. Examination of the injection site 24 hours after administration revealed heavy deposits of subtilin, and a blood assay 1 month after injection showed a trace of subtilin, indicating that absorption was still in progress. Intravenous administration produces blood levels of some magnitude. A dose of 10 mg./kg., given as a 1% solution in 5% glucose in the ear vein of rabbits, gives a blood concentration of approximately 100–200 ppm. in 5 minutes, dropping to 10–30 ppm. in 2 hours and to zero in 24 hours. Greater dosage may kill by interfering with circulation between heart and lungs, presumably by embolism. The intravenous LD_{50} for rabbits is not known accurately; for mice, it is about 100 mg./kg. (Chin). Intravenous infusion (20 mg./kg./hr.) was given a rabbit for 4 hours without producing visible symptoms, and leading to a final blood level of about 750 ppm.

The effect of antu on the body temperature in the rat. MARTIN M. WINBURY and JOHN LOVE (introduced by W. E. Hambourger). *From Lehigh Valley Laboratories, Easton, Pennsylvania.* In rats a single oral dose of ANTU (α -naphthylthiourea) provokes a rapid fall in body temperature which frequently begins as early as 15 minutes after administration. Within 9 hours following a sublethal dose (0.94 to 1.88 mg. per 100 gm.) the temperature is reduced about 2.5°C., returning to normal 3 to 5 hours later. On administration of a lethal dose of ANTU (3.75 to 15 mg. per 100 gm.), the body temperature falls rapidly during the first 4 hours from the control of 37.4 ± 0.14°C. to about 33°C. and then more gradually to the low of 29.6 ± 0.51°C. at death. It has been well established that the rat will develop a tolerance to toxic doses of ANTU by previous administration of this substance (Richter, C.P., J. Am. Med. Assn., 128, 927, 1945). We find that a single sublethal dose not only affords protection against the lethal effects of a toxic dose but also prevents or reduces its hypothermic action.

Effect of ouabain and digoxin on the energy-rich phosphate store of the heart. ALBERT WOLLENBERGER (introduced by OTTO KRAYER). *Harvard Medical School.* Ouabain and digoxin, administered to the dog heart-lung preparation, do not cause significant changes in the phosphocreatine and adenosine polyphosphate (APP) content of the heart, as long as their action is mainly inotropic. The toxic action of these compounds, on the other hand, is associated with a progressive loss of phosphocreatine. At the onset of ventricular fibrillation, this loss amounts to about 75 per cent. The APP concentration in the poisoned hearts is not lowered more than can probably be accounted for by cardiac edema. Both poisoned and unpoisoned hearts contain only insignificant amounts of acyl phosphate. Theoretically, the observed decrease in phosphocreatine reflects changes in the turnover of the labile phosphate groups of APP. It can be explained in three ways: (1) Synthesis of APP may be retarded. Presumably this would be the result of a depression of oxidative metabolism which toxic concentrations of cardiac glycosides are known to produce in the myocardium. (2) Breakdown of APP may be accelerated. This might take place also during the non-toxic stage, but might not be apparent because of increased phosphorylation. (3) Both factors 1 and 2 may be operating.

The effect of splenectomy upon the susceptibility of mice to infection by trypanosoma cruzi. ORLYN WOOD (by invitation) DORIS NOSHOLD (by invitation) and LLOYD D. SEAGER. *Department of Pharmacology, Woman's Medical College of Penna.* Splenectomized and normal mice were inoculated intraperitoneally with approximately 50,000 organisms per kg. In one series of experiments a Mexican strain of T. cruzi was used and in a second series a more virulent Brazilian strain was employed. One group of 20 splenectomized mice were inoculated within a few days of the operation. In these the peak of infection was reached more slowly and mortality was lower than in the controls. In the rest of the experiments (120 mice) 10 to 20 days were allowed to elapse post operatively before inoculation. In these the peak of infection was reached more rapidly and the mortality was higher than in the controls. It is suggested that the inflammatory reaction from the operative procedure made the peritoneal cavity an unsuitable environment for the organism for a few days following splenectomy.

Effects observed in dogs following the prolonged feeding of DDT and its analogues. GEOFFREY

WOODARD (by invitation), BERNARD DAVIDOW (by invitation), and ARTHUR A. NELSON. *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington 25, D. C.* The o,p′ and p,p′ isomers of DDT, technical DDT, the dehydrochloride of DDT, DDD, and the dimethoxy analogues of DDT (DMDT) were fed to dogs for varying periods of time up to 49 months in some cases. The insecticides were administered daily except Sunday in oil solutions contained in gelatin capusles. Undissolved crystalline DDT was fed in one experiment. Chemical analyses of samples of fat taken at yearly intervals from the same dogs fed various levels of DDT showed accumulation to increase with dosage, time, and administration in solution. DDD also accumulated in fatty tissues. The average survival times in days at an 80 mg./kg./day level of intake are: p,p′-DDT, 55; o,p′-DDT, 37; technical DDT, 55; DDD, 80; and DDT dehydrochloride, above 120. Dogs fed DMDT at 330 mg./kg./day still survive after 120 days. At the 80 mg./kg./day levels of o,p′-DDT, p,p′-DDT and technical DDT, histopathological examination showed the major effect produced by all three substances was moderate to severe liver damage made up of central atrophy or low grade necrosis, subacute hepatitis, and fatty degeneration. The same level of DDD produced slightly less liver damage and no jaundice or hemorrhages but did cause a marked atrophy of the adrenal cortex. No significant pathologic changes were noted in the DDT dehydrochloride animals.

Oxytocic drugs and dysmenorrhea. R. A. WOODBURY, GEORGE P. CHILD (by invitation), and R. TORPIN (by invitation). *Division of Pharmacology, University of Tennessee, Memphis, and Departments of Pharmacology and Obstetrics and Gynecology, University of Georgia School of Medicine, Augusta.* Satisfactory relief was obtained in thirty-two of thirty-four dysmenorrhea patients by subcutaneous administrations of pitocin; in the remaining two the relief was incomplete. The beneficial effects included removal of extra-uterine distress such as menstrual tension and headache as well as the abdominal distress.

Experiments were designed to investigate the influence of pitocin on the uterus in humans. Using the four balloon technique it was observed that pitocin increased the peristaltic effects of the uterine activity by bringing about better relaxation of the lower uterine segment between contractions and greater pressures during contractions. Previously it has been reported that under certain conditions pitocin caused dilation of the uterine vascular bed of dogs. Apparently the beneficial effects of pitocin can be explained partly by the rhythmic uterine contractions and partly by the reduced spasm of blood vessels. Rhythmic uterine contractions facilitate blood flow to the uterus and expel debris.

Similar studies with posterior pituitary solution, pitressin and ergonovine demonstrate that these drugs tend to increase the frequency and amplitude of contraction in all parts of the uterus including the lower segments. Apparently these drugs do not bring about the same type of uterine activity as is elicited with pitocin. In addition pitressin produces vasoconstriction which in turn may contribute to the fact that injections of pitressin have elicited dysmenorrhea-like distress in forty-four of forty-five patients.

These observations that dysmenorrhea distress may be relieved by a drug which facilitates rhythmic uterine activity should broaden the search for effective therapeutic agents for the cyclic distress.

Rosaniline base (CI 677) as a prophylactic in Schistosoma mansoni infections in mice. HAROLD N. WRIGHT, ELIZABETH M. CRANSTON, WAYNE A. CHADBOURN (by invitation), ASHTON C. CUCKLER (by invitation), DOMINIC DEGUISTI (by invitation) and RAYMOND N. BIETER. *Department of Pharmacology, University of Minnesota, Medical School, Minneapolis, Minnesota,* Mice were infected intraperitoneally with 100 to 125 cercariae of Schistosoma mansoni obtained from snails of the species Australorbis glabratus. Therapy of infected mice with rosaniline base, using the drug-diet method of administration, was started at various time intervals before or after infection and continued for one to four weeks. Mice were autopsied between eight and twelve weeks after infection, at which time the number of worms in the portal and mesenteric veins and liver were counted and the relative degree of egg deposition in the liver noted. When therapy with rosaniline base in a concentration of 0.33 per cent in the diet was started one week before infection and continued for twenty-eight days no worms or eggs were found at autopsy in 90 per cent of the mice, whereas only 13 per cent of control mice were negative. With similar therapy begun two weeks after infection 80 per cent of mice were negative for worms and eggs. Shorter periods of therapy resulted in progressively decreasing activity. When therapy was withheld for more than two weeks after infection only a slight therapeutic action was obtained regardless of the length of therapy. From the results obtained rosaniline base appears to possess a high degree of prophylactic activity and a low degree of curative activity in Schistosoma mansoni infections in mice. The possible activity of other triphenyl methane dyes is under investigation. This work was done under contract with the Office of the Surgeon General of the United States Army, the United States Public Health Service and the University of Minnesota.

Comparison of biological and chemical evalua-

tions of sex hormone balance. M. X. ZARROW (by invitation) and W. T. SALTER. *From the Laborae tories of Pharmacology and Toxicology, Yal- University of Medicine, New Haven, Connecticut.* In studying the urinary excretion of sex steroids chemical methods are available which include degradation products in addition to the original hormone. These degradation products possess varying degrees of residual biological activity which is not indicated in the chemical analysis. It is a moot point whether these break-down products should be rated at their biological equivalents or in accord with the original chemical origin. Accordingly studies have been made on the urine of normal human subjects and patients suffering from cancer (mammary and prostatic) and various endocrinopathies such as gynecomastia, hypogonadism, adrenal virilism and eunuchoidism. Biological determinations were carried out on urine samples hydrolyzed with HCl and extracted with ether. The "estroids" were assayed in castrated mice using estrone as a standard and the androgens were assayed in day-old cockerels using androsterone as a standard. Chemical determinations were made by the technique of Cahen and Salter (J. Biol. Chem. **152:** 489, 1944) for 17-ketosteroids and of Oesterling and Salter (Fed. Proc. **6:** 171, 1947) for "estroids." The results from the chemical determinations tend to give higher values than obtained by biological assay. For example, in pooled urine from a group of soldiers with nutritional gynecomastia the chemical value for "estroids" was 8 micrograms per 24 hours as compared with 4 micrograms obtained by bioassay. Howeve:, in two cases with negative values for 24 hour determination in urinary estrogens by the chemical method negative results were also obtained in the bioassay. More detailed data in various pathological conditions are reported.

The effect of tetraethyl ammonium chloride on gastric secretion and motility. MITCHELL ZWEIG (by invitation), F. STEIGMANN, and KARL A. MEYER (by invitation). *Hektoen Institute for Medical Research of Cook County Hospital and the Department of Surgery, Northwestern University, School of Medicine.* Tetraethyl Ammonium Chloride (Etamon: Parke, Davis Co.), which has the ability to block at the autonomic ganglia the transmission of both sympathetic and parasympathetic nerve impulses, was employed in a preoperative study of 11 patients with hypertension for determination of results which might be expected following sympathectomy. Because of paucity of reports on the effect of this drug on gastric functions, it was considered pertinent to study its effect on gastric motility and secretion. The following procedure was used: A balloon was passed into the fundus of the stomach, inflated with 10 cc. of air, and then attached to a kymograph. Gastric juice was aspirated through a Levine tube. The total quantity aspirated in fifteen minutes was measured and titrated for both free and total acid. A base level of secretion and motility was taken for at least one hour. After obtaining the base level, 400 to 600 mgm. of Tetraethyl Ammonium Chloride were given intravenously. Within five minutes after the injection of the drug every patient had a complete cessation of motility. This lasted from 45 to 100 minutes. The amount of free acid in milliequivalents was determined for an equal period before and after the Tetraethyl Ammonium Chloride was given. In every instance, except in four patients who secreted no free acid during the test period, the acid levels were reduced after the drug was given.

THE AMERICAN SOCIETY FOR EXPERIMENTAL PATHOLOGY

THIRTY-THIRD ANNUAL MEETING

Atlantic City, New Jersey, March 15, 16, 17, 18, 19, 1948

(For possible corrections in any of the following abstracts see the next issue).

The transparent chamber technique in the mouse in the study of tumor histo-physiology (Motion Picture). GLENN H. ALGIRE and FRANCES Y. LEGALLAIS (by invitation). *National Cancer Institute, National Institute of Health, United States Public Health Service, Bethesda, Md.* The transparent chamber technique permits in vivo microscopic observation of growth and vascular development of transplanted tumors over intervals up to 100 days. Examination and photography of blood vessels and cells are carried out at magnifications up to 1000X, using oil immersion objectives. The method is being applied to problems of tumor induction and growth, effect of chemical and physical agents on tumors, cell migration and division, immunity, and circulatory physiology in normal and neoplastic tissues. The motion picture illustrates the procedures for operation and examination of the living tissues.

Discrepant animal and clinical observations on

hypothermia and procaine. FREDERICK M. ALLEN. *City Hospital, Welfare Island, New York, N. Y.* Different temperature reactions of different species result in discrepancies regarding treatment of conditions such as burns and visceral shock, but reduced temperature is supported by the general evidence. Laboratory animals react to procaine differently than man to such an extent that there is doubt of adequate duplication of the recent clinical uses of this drug, on a physiological basis. The new concepts of certain pathological processes may be still more difficult to elucidate in the laboratory.

The relationship of caloric intake level and protein intake level to rate of protein synthesis. EARL P. BENDITT. *Department of Pathology, The University of Chicago Clinics, Chicago.* The purpose of these experments was to determine the influence of caloric and protein intake levels on the rate of tissue synthesis in protein depleted animals. Standard protein depleted animals were used. The method of protein depletion and repletion have been described (Wissler, Woolridge, Steffee and Cannon, J. Immunol. 52: 267, 1946). Caloric intake was varied at constant protein intake; protein intake was varied at constant caloric intake and the two were varied together. The protein was lactalbumin and casein (half and half) and the rations were adequate in minerals, vitamins, roughage and fats. Five or six animals in individual cages were fed each ration for a period of 14 days. They were then sacrificed and the carcasses analyzed for protein, fat, water and ash. The intakes were calculated as Cal./m²/day and gm. of protein/kgm. body weight/day, and rates of protein fabrication as gm. protein gained/kgm. body weight/day. The results were as follows: 1) Limitation of caloric intake below 1240 Cal./m²/day restricted protein utilization. 2) Caloric intakes above this level did not augment protein synthesis, but were associated with increased deposition of fat. 3) With an adequate caloric intake the rate of protein utilization increased with rising levels of protein intake over the range studied (up to 14.6 gm/kgm/day). 4) Growing and mature protein-depleted animals were found to react in an almost identical fashion.

Production of intimal atherosclerosis by intravenous injection of colloidal cholesterol into rabbits. MARGARET BEVANS, LIESE LEWIS ABELL, and FORREST E. KENDALL (introduced by H. P. SMITH). *From the Research Service, First (Columbia) Division, Goldwater Memorial Hospital, Department of Hospitals, City of New York, and the Department of Medicine, College of Physicians and Surgeons, Columbia University.* Colloidal solutions containing 2.5% cholesterol stabilized with 1% sodium stearate can be rapidly injected into the ear vein of rabbits without producing adverse symptoms. The injection of 0.5 gm. of cholesterol into a 2.5 kg. rabbit leads to an increase in the serum cholesterol level amounting to 250–300 mg.% above the initial concentration 10 minutes after injection. The level falls to a value 100–150mg. above the base line in 6 hours and then rises again to 150–200 mg. at the end of 24 hours. Normal levels are regained in from 4 to 7 days. Daily injections of cholesterol result in serum levels comparable to those obtained when the same amount of cholesterol is fed and lead to arterial lesions indistinguishable from those obtained in feeding experiments. Gross intimal lesions were first noted after the injection of 6.5 gms. of cholesterol over a period of 13 days. These were few and small and occurred only in the ascending aorta. Extensive lesions were produced by the injection of 14.5 gms. of cholesterol over a period of 36 days.

Cholesterol in intestinal and hepatic lymph of the rat. JESSE L. BOLLMAN and EUNICE V. FLOCK. *Division of Experimental Medicine, Mayo Foundation, Rochester, Minnesota.* Intestinal lymph was drained continuously through a small polyvinyl plastic tube from the main intestinal lymphatic near the cysterna chyli. After a control period the rats were fed by stomach tube on the second and second and third day. Samples of lymph were collected to cover periods of 3, 6, 9, 12 and 24 hours after each meal. Following a fat free meal to which cholesterol was added the concentration of cholesterol of the lymph was increased over the control values for about 12 hours and usually returned to the control values before 24 hours. Both free and ester cholesterol increased but the greater increase was that of cholesterol esters, although considerable variation in the relative proportion of each was found. Addition of oleic acid or of neutral fat to the same diet containing cholesterol did not greatly increase the concentration of cholesterol appearing in the lymph. The amount of cholesterol absorbed via the lymph is extremely small compared to the lymphatic absorption of fat. The concentration of cholesterol and of cholesterol esters in hepatic lymph is similar to that of plasma and the fluctuations are much smaller than in intestinal lymph.

Blood iodine in alloxan diabetes of dogs. H. G. DAVIS, JR. (by invitation) and R. D. BAKER. *Department of Pathology, The Medical College of Alabama.* Total blood iodine studies were done on two groups of eight dogs using Steven's acid-ashing method. Six of each group were made temporarily or permanently diabetic with alloxan. One group received food containing 0.5 mgm. of iodine daily. The other did not. Two dogs of each group were retained as controls. In every case a rise in blood iodine was obtained where there was sufficient alloxan dosage to cause the blood-sugar to rise. The blood iodine remained high in the dogs which became permanently diabetic. The values for those animals receiving iodine supplement rose considerably higher than those which did not. Theories suggested to explain the rise in blood iodine include: (1) Damage to the kidneys by alloxan with

failure of excretion. (2) Damage to the liver with failure of hepatic excretion of iodine into the intestine. (3) Removal of endocrine action of islet tissue on the thyroid, allowing excessive excretion of iodine by the thyroid.

Iron absorption in normal subjects and in patients with anemias of varied etiology. REUBENIA DUBACH (by invitation), SHEILA CALLENDER (by invitation) and CARL V. MOORE. *Department of Internal Medicine, Washington University School of Medicine, St. Louis, Mo.* Measurement of iron absorption by the isotope technic has proved difficult because not all of the iron absorbed from an oral dose may be utilized for hemoglobin formation. It has become necessary, therefore, to alter the procedure so that retention could be divorced from utilization. Accordingly, the method has been extended to include determination of the unabsorbed radioiron in feces as well as that converted into hemoglobin. Recovery of radioiron from feces was accurate within 10%. The test dose equalled 1mg./kg. body weight and was given as ferrous chloride. In iron-deficient subjects, all of the radioiron not recovered in the feces was utilized promptly to form hemoglobin. Results in normal persons indicated that a small amount of absorbed iron was not converted into hemoglobin; the amount recovered in feces plus that found in blood equalled 83 to 100% of the test dose. In 12 subjects with fever, untreated pernicious anemia, refractory anemia, and hemolytic anemia there was a wider discrepancy between the amount of iron retained and the amount utilized, and some of these patients absorbed a relatively large amount of the metal. For instance, 2 patients with Hodgkin's disease and an associated hypochromic anemia retained 52 and 32%, respectively, yet utilized only 15 and 2.5%. In 3 patients with untreated pernicious anemia, the proportion of retained iron which appeared in the blood was small at first, but increased sharply after liver therapy. Fecal recoveries indicated that they had absorbed from 15 to 33% of the test dose. From these results, it is concluded that: 1) the per cent of a test dose of radioiron which appears as circulating hemoglobin should not be accepted as a measure of the total amount absorbed except in afebrile patients with hypochromic anemia; and 2) if iron absorption is regulated by the degree of "block" present in nucosal cells of the intestinal tract, as seems to be the case, the block is only relative and is not nearly as efficient or complete as earlier work had indicated.

A study of experimental serum sickness. WILLIAM E. EHRICH, CAROLYN FORMAN (by invitation) and JOSEPH SEIFTER. *Philadelphia General Hospital, University of Pennsylvania, Wyeth Institute Applied Biochemistry.* Serum sickness was produced in rabbits by injecting them with horse or duck serum. It was confirmed that large doses of salicylates inhibit the production of certain phenomena of this condition. As these doses caused both an alarm reaction and an anticoagulant effect, the role of these phenomena in serum sickness was investigated by treating serum injected rabbits with alarming stimuli such as colchicine, or with anticoagulants such as dicoumarol or algarin (a heparin-like compound). The results of these studies will be presented.

Respiration of tissues from hypersensitive animals when antigen is added in vitro. RICHARD H. FOLLIS, JR. *Department of Pathology, Johns Hopkins University.* The anaphylactic type of hypersensitivity (Arthus phenomenon, protein-type hypersensitivity) is characterized by contraction of smooth muscle and damage to endothelium of blood vessels. Whether the local Arthus reaction which can be elicited in heart, kidney, testis, etc. is due entirely to vascular damage is not clear. Biochemical studies of isolated tissues would seem to offer an approach to this problem. Guinea pigs and rabbits were injected respectively with egg albumin and horse serum. At appropriate intervals the animals were tested intracutaneously with antigen. When they were found to exhibit a strong Arthus phenomenon, the oxygen consumption of various tissues: liver, kidney, smooth muscle, heart muscle and testis, was then determined manometrically using Krebs-Ringer solution with or without added homologus serum as substrate. Specific antigen or control material was added from the side arm after equilibration and hourly readings made up to six hours. Experimental data indicate that there is no appreciable change in the oxygen consumption (in micro liters) per mg. tissue (dry weight) per hour from normal when specific antigen is brought in contact with cells of the organs tested. Such findings would confirm and extend observations heretofore recorded

Histochemical studies of basement membranes. ISIDORE GERSH. *Department of Pathology, University of Illinois College of Medicine, Chicago.* Basement membranes in skin, developing kidney, thyroid gland, and lung of normal organs have been studied after staining for reticular fibers and for an optically homogeneous component. The latter has been found to be a carbohydrate-containing protein, which can be dissolved or destroyed by certain reagents. These also affect the "ground substance" of the connective tissue in a similar way. The "ground substance" and the homogeneous component of the basement membrane are morphologically continuous. During histogenesis, as well as in inflammation, morphological changes take place in the homogeneous component which suggest that the basement membrane may be a highly labile, modifiable structure.

Sodium-potassium levels and adrenal gland necrosis in experimental simian malaria. ALFRED

GOLDEN and R. R. OVERMAN (by invitation). *Departments of Pathology and Physiology, University of Tennessee, College of Medicine, Memphis, Tennessee.* Monkeys experimentally infected with P. knowlesi show an average survival when untreated of five days. During their illness they develop profound alterations of blood potassium and sodium levels as determined by flame photometry. Associated with these and other biochemical changes there is consistent focal necrosis of the adrenal gland cortex which varies from focal lesions in all zones of the cortex to large patches of such necrosis associated with hemorrhage. An entirely similar lesion was demonstrated at autopsy in a human case of P. vivax infection.

Adrenal gland lesions in experimental simian malaria and similar human lesions in varied diseases. ALFRED GOLDEN. *Department of Pathology, University of Tennessee, College of Medicine, Memphis, Tennessee.* Lesions entirely analagous to those seen in the adrenal glands of monkeys dying with acute P. knowlesi infection were observed in the following diseases at post mortem examination: B. pyocyaneus endocarditis, healing impetigo and fatal diarrhea in a newborn infant; disseminated tuberculosis in an adult; acute meningococcic meningitis and septicemia; mutliple gunshot wounds with multiple perforations of the gastrointestinal tract; acute gangrenous cystitis and acute gangrenous pyelonephritis; multiple abscesses of the kidney and lung; fulminant hypertension with terminal bronchopneumonia; severe body burns; acute P. vivax malaria in an adult; gangrenous vaginitis following radium implantation for a carcinoma of the cervix; acute interstitial nephritis. The evolution of the lesion can be traced from its minimal manifestations to its severest forms. The lesion in identical to that seen previously by the author in epidemic typhus fever.

Susceptibility of macacus cynomolgus to Japanese encephalitis virus with special reference to the alimentary tract. F. B. GORDON, F. M. SCHABEL, JR. (by invitation) and MARGARET ABENDROTH (by invitation). *Department of Bacteriology and Parasitology, University of Chicago.* As part of a series of experiments on the susceptibility of various animals to infection by feeding with neurotropic viruses, 2 cynomolgus monkeys were fed bread soaked in an emulsion of Japanese encephalitis virus. No clinical abnormalities resulted but one of the monkeys had a viremia from the 3rd to the 10th day. No virus could be recovered from oral or rectal swabs by subinoculation of mice with penicillin-treated suspensions. The monkey with viremia resisted a later intracerebral inoculation of virus (18,000 mouse infectious doses) which produced severe encephalitis in the other fed monkey and two controls. In the 3 latter monkeys, infected by intracerebral injection, viremia occurred early but terminated on the fourth to the 7th day, but the time of the onset of illness. Oral and rectal swabs were tested for virus at intervals between the time of inoculation and death. No virus was found in the rectal swabs, but it was demonstrated once in small amount in the mouth on the 5th day after intracerebral inoculation. The 3 monkeys were killed when severely affected, and some 18 tissues of each were tested for virus. It was recovered from the central and peripheral nervous systems, from leg muscle, and from the anterior end of the alimentary canal, but none was found in the intestine.

Failure of trypanosoma cruzi lysate in treatment of Brown-Pierce carcinoma of rabbit. O. M. GRUHZIT and R. A. FISKEN (by invitation). *Research Laboratories, Parke, Davis, & Co., Detroit, Mich.* A non-virulent Trypanosoma cruzi strain was grown on beef blood in heart-brain broth effusion agar medium for fourteen to sixteen days. The trypanosomes were harvested by centrifugalization, yielding a sediment of five billion organisms per cubic centimeter. A distilled water lysate, 1:50 with 1:5,000 dilution of cetyl trimethyl ammonium bromide, each cubic centimeter of which contained about 100 million organisms, was prepared from a 72 hour T. cruzi harvest, kept at −6°C. A group of nine rabbits was inoculated intratesticularly with Brown-Pierce rabbit carcinoma cell suspension. Three days after inoculation, five animals received intramuscularly 1.0 cubic centimeter each twice daily for seventeen days of the T. cruzi lystate. In eight days, the treated and untreated rabbits showed nodular growth in the testes. On the tenth day, the consolidated testis of one treated rabbit was excised and part of the emulsified material was injected into the testis of each of four normal rabbits, which eight days later showed neoplastic growth. At the end of seventeen days of treatment with the lysate, the treated and untreated rabbits presented extensive nodular consolidation of the testes. On the twenty-seventh day, three treated rabbits showed neoplastic metastasis in one or both eyes, including the one from which the tumor-bearing testis was removed, but none occurred in the control rabbits. The treated and two control rabbits showed massive tumor growths in the testes. In the other two control rabbits, the tumors had regressed. Under our conditions, the use of T. cruzi lysate had no inhibitory effect on the growth of the Brown-Pierce carcinoma in rabbits.

Tumor therapy by the direct infiltration of radioactive colloidal metallic gold. P. F. HAHN. *Vanderbilt University School of Med., Nashville, Tenn.* If the list of available radioactive isotopes is carefully considered from a standpoint of potentially useful therapeutic agents, keeping certain logical criteria in mind, it is found that there are probably less than ten which would be suitable. These

criteria include: 1. Desirable radiation spectrum; 2. Known biological behaviour; 3. Suitable half-life; 4. Known chemical behaviour; 5. Good cross-section for bombarding particles used; 6. Absence of long-lived contaminants; and 7. Economical production. Gold^{198} made in the chain reacting uranium pile by absorption of slow neutrons by the 100% abundant isotope Au^{197} appears to meet these criteria to a degree not approached by other isotopes. In addition it has specific properties such as its metallic colloid's insolubility in all body tissue fluids which render it particularly valuable as a therapeutic agent. By means of a needle of fine calibre this material may be infiltrated directly and throughout a tumor mass in the same manner one would use novocaine. It has been found that when administered in this manner the colloid remains at the site of infiltration. Whereas in the use of radium needles and radon seeds it is necessary to filter out the alpha and beta radiation in order to prevent sloughing at the site of application due to over-intense localized radiation effects, and to rely only on the gamma radiation, using the gold isotope one makes use of the beta radiation from not six or eight sources but from many billions of point sources. Thus the ionizing radiation is delivered uniformly throughout the desired mass of tissue and adjacent structures are unaffected since the mean path of the beta particles is a millimeter or less. Results of clinical application will be presented.

The renal lesion of murine haemobartonellosis compared with lower nephrone nephrosis. ROBERT S. HAUKOHL (introduced by W. A. D. ANDERSON). *Department of Pathology and Bacteriology, Marquette University School of Medicine, Milwaukee, Wis.* The renal changes were studied in 34 rats dying in the acute stage of haemobartonellosis following splenectomy and the inoculation of infected blood. The renal lesion is characterized by: 1) marked degeneration, necrosis and desquamation of the epithelium of the distal convoluted tubules, 2) accumulations of hyaline droplets in the epithelium of the distal convoluted tubules, 3) precipitation of heme pigment casts and crystals in the tubules of the lower nephron and collecting tubules, and 4) minimal structural change of the glomeruli, upper and intermediate nephrons and interstitial tissue. The heme pigment casts and crystals and the hyaline droplets stain positively with the hemoglobin stain of Dunn and Thompson (Arch. Path. **39**: 49, 1945), and fail to stain with Prussian blue. Two factors are essential for the production of this lesion: 1) relative renal anoxia secondary to shock, anemia and the vaso-constrictor action of circulating free hemoglobin, and 2) the excretion of large amounts of hemoglobin with precipitation in the lumen (casts and crystals) and reabsorption by the tubular epithelium (hyaline droplets). The renal lesion of haemobratonellosis is essentially similar to the renal changes in human lower nephron nephrosis. The appearance of hemoglobin positive hyaline droplets in rat kidneys and not in the human counterpart may be attributed to the relatively large amount of hemoglobin excreted in haemobartonellosis. The absence of interstitial reaction is due to the relatively short interval between the onset of hemoglobinuria and death.

Protection against bacterial endotoxin by penicillin and its impurities. WALTER D. HAWK (by invitation) and C. PHILLIP MILLER. *Department of Medicine, The University of Chicago, Chicago, Illinois.* The protective action of pure penicillin against a number of bacterial endotoxins, already reported, has been increased by the addition of certain substances present in impure preparations of penicillin. Mice were treated with three intraperitoneal injections of 0.5 ml. of the penicillin preparation during the 24 hours preceding intraperitoneal injection of bacterial endotoxin (S. Aertrycke). About 10,000 units of penicillin per mouse were used. Untreated animals received physiological saline and other control solutions before their injections of endotoxin. Treatment with crystalline penicillin increased the LD50 of endotoxin to about twice that obtained in controls. Treatment with the impure penicillin increased the LD50 to more than six times that obtained in the controls. Treatment with the impurity, unaccompanied by penicillin, provided no protection against the endotoxin. The effective impurity is water soluble, filtrable, heat-stable, resistant to acidification, and soluble in certain organic solvents.

Influence of various protective compounds in preventing fatty changes by dichloroethane. BENJAMIN HIGHMAN. *National Institute of Health, Pathology Laboratory.* Several series of white male rats of the Sprague Dawley strain, weighing approximately 90 grams, were injected subcutaneously with 1 ml./kg. of undiluted dichloroethane. The mortality rate was low, but the rats developed severe though transitory fatty degeneration of the liver, kidney and heart, as shown by frozen sections stained for fat. These changes were usually marked in rats killed 24 hours after the injection. When 1000 mg./kg. of dl-methionine in 2 per cent aqueous solution was injected intraperitoneally immediately after the dichloroethane, these changes were much less marked and were minimal or absent when an additional 500 mg./kg. of methionine was injected 5 hours later. The results were reproducible. Larger rats were relatively more susceptible to dichloroethane and diluting the compound with oil increased its toxicity often leading to considerable mortality even after methionine administration. Various agents

were tested, using as controls groups of unprotected and methionine protected rats simultaneously injected with dichloroethane. No compounds tested equaled methionine in potency. Slight protection was offered by some compounds containing an amino group in a benzene ring like sulfanilamide (given by stomach tube) and aniline. Usually the heart was most readily protected and the liver least. The results are consistent with previous findings based on the ability of such compounds to reduce mortality. It is suggested that such histologic methods be used to supplement other methods for testing the efficacy and mode of action of various substances for protection against other toxic agents producing transient histologic changes.

Arterial disease may be a matter of days, not decades. RUSSELL L. HOLMAN. *Department of Pathology, L. S. U. School of Medicine, New Orleans.* There is a growing body of evidence incriminating *certain lipid substances under certain conditions* as an important factor in the pathogenesis of arterial disease. This applies regardless of species, but the form the arterial lesions take varies widely in different species and is apparently conditioned by the pattern of lipid metabolism characteristic of the species. For this reason it seems propitious at this time to shift emphasis from anatomical form to some of the broader aspects of pathological physiology that may be involved in the genesis of arterial disease. Data previously presented to this society have shown that arterial lesions can be produced with regularity in dogs by feeding a specified high fat diet for two months or longer, than damaging their kidneys in any of several ways. These lesions, which constitute another example of arterial disease related to a disturbance in lipid metabolism, have been observed as early as four days after the production of renal insufficiency. These studies have suggested that arterial disease may be a matter of days, not decades. Several human cases consistent with this suggestion are presented, and some of the implications and possible applications of the suggestion are discussed.

Failure of implantation in vitamin E deficient rats. HANS KAUNITZ and CHARLES A. SLANETZ (by invitation). *From the Departments of Pathology and Animal Care, College of Physicians and Surgeons, Columbia University, New York City.* A colony of highly inbred rats was kept for seven generations on a purified vitamin E deficient diet containing 10 per cent lard. Adult animals therefore consumed roughly 30 mcg. tocopherol daily because lard was the only measureable source of tocopherol. The controls obtained the same ration supplemented by 3 mcg. synthetic dl alpha tocopherol, (Hoffmann La Roche) per 100 gms. diet. 721 mating experiments, carried out between the ages of 5 weeks and 20 months, revealed that after the 8th week the rate of implantation in the deficient group was significantly lower (chi square) than in the controls. The differences persisted when the body weight of the animals was taken into account. It seems that there exists a specific tocopherol requirement for implantation steeply increasing with age. A daily tocopherol intake of roughly 30 mcg. (as provided by the basic ration) enabled 50 per cent of the 2–4 months old animals to become pregnant. The requirements for implantation represent only a fraction of those necessary for normal gestation and lactation. Experiments are being performed which may clarify whether the implantation failure in vitamin E deficiency is caused by ovarian or uterine dysfunction or by disturbances of the fertilization or the passage of the ovum from the ovaries to the uterus.

Effect of intravenous injection of oxidized cholesterol upon the production of atherosclerosis in rabbits. FORREST E. KENDALL, WALTER MEYER, and MARGARET BEVANS (introduced by H. P. SMITH). *From the Research Service, First (Columbia) Division, Goldwater Memorial Hospital, Department of Hospitals, City of New York, and the Department of Medicine, College of Psysicians and Surgeons, Columbia University.* Bergstrom and Wintersteiner (J. Biol. Chem. **141**, 597, 1941) showed that colloidal solutions of cholesterol are readily oxidized by air to give 7-ketocholesterol and 7-hydroxy cholesterol as the principal products. Intravenous injection of these oxidized solutions into rabbits leads to blood cholesterol levels comparable to those obtained with the unoxidized preparations. However striking differences are found in the immediate effect of the two preparations upon the arterial walls. Lipid droplets can be detected within the cells of the intima 24 hours after a single injection of 0.5 gms. of oxidized cholesterol. 72 hours later proliferation of the intimal cells can be seen. These lesions persist without much change for at least 6 months. Multiple injections lead to an immediate increase in the amount of sudanophilic material in the intima which roughly parallels the amount of oxidized cholesterol injected. These early changes are not produced by the injection of unoxidized cholesterol. However if high serum cholesterol levels are maintained for several weeks by the injection of either material there is little difference in the extent or character of the lesions produced.

The production of phage in the absence of cellular growth. A. P. KRUEGER, T. COHN (by invitation) and P. N. SMITH (by invitation). *From the Department of Bacteriology and Office of Naval Research Task V, University of California, Berkeley, California.* Reproduction of the cellular sub-

strate is generally considered to be an essential condition for the formation of phage although some evidence has been secured indicating that the two process may be separated. We have described elsewhere the accelerating action of penicillin on phage-engendered lysis of Staphylococci and in the present experiments, we have utilized this effect to impose conditions such that [phage] increases without detectable bacterial growth. Actively growing Staphylococci are centrifuged from the medium and are resuspended in Locke's solution, at 5°C., containing 1.0 unit of penicillin G/ml. after 1.0 hour, phage is added to the suspension using a volume which does not introduce a significant amount of nutrients. The mixture is kept an additional 1.0 hour at 5°C. and is then shaken in a water bath at 36°C. [bacteria] is determined at brief intervals throughout the reaction by means of direct microscopic counts and nephelometric measurements in the Klett-Summerson photoelectric comparator. After lysis is complete (about 1.5 hours after removing the suspension to the 36° bath) [phage] is determined by Gratia's plaque count method. Starting with 1×10^8 plaques/ml., the titre after lysis is found to be between 4×10^8 and 5×10^8 plaques/ml. [bacteria] remains constant until massive lysis begins about 0.6 hours after bringing the suspension to 36°C.

The effect of bacterial endotoxins on the carbohydrate metabolism of the rabbit. ERNEST KUN (introduced by C. PHILLIP MILLER). *Departments of Pharmacology and Medicine, University of Chicago, Chicago.* Rabbits were injected intravenously with a dose of meningococcal or Salmonella endotoxin, which caused death in 2–3 hours. The blood glucose showed a rapid increase, followed by hypoglycemia. Blood inorganic phosphate and lactic acid increased, while blood pyruvic acid diminished. There was a marked decrease in liver and muscle glycogen. Liver and muscle lactic acid increased, while pyruvic acid diminished. Succinic dehydrogenase was significantly inhibited in both liver and muscle. Cytochrome oxidase was not affected. (Supported by a grant from the U. S. Navy, Office of Naval Research.)

Plasma tocopherol levels in cardiac patients. JANET M. LEMLEY (by invitation), ROBERT G. GALE (by invitation), ROBERT H. FURMAN (by invitation), MARY E. CHARRINGTON (by invitation) and GEORGE R. MENEELY. *From the Departments of Medicine and Biochemistry of Vanderbilt University School of Medicine.* The importance of tocopherol in maintaining normal muscular function has been established. Cardiac manifestations have been described in vitamin E deficient animals. Claims have been made of benefit to cardiac patients following tocopherol administration despite lack of evidence that vitamin E deficiency exists in humans. Twenty five cardiac patients were studied for evidences of tocopherol deficiency. The tocopherol content of the blood plasma was determined by the method of Quaife and Harris. Normal values by this method average 1.15 mg. per 100 ml. of plasma. The lower limit of normal is not firmly established. Some consider 0.88 mg. % as this limit, while others place the lower limit of normal at 0.64. The mean plasma tocopherol level for the group studied was lower than the normal mean. Several patients exhibited sufficiently low plasma tocopherol levels to suggest deficiency. The evidence presented does not indicate a general deficiency of tocopherol in cardiac patients but suggests that in some patients with heart disease tocopherol deficiency might be a factor.

On the demonstration of hyaluronidase in cercariae of schistosoma mansoni. MILTON D. LEVINE, RAY F. GARZOLI, ROBERT E. KUNTZ and JOHN H. KILLOUGH, (introduced by K. M. ENDICOTT). *Naval Medical Research Institute, National Naval Medical Center, Bethesda, Maryland.* Hyaluronidase in relatively high concentration has been shown to be present in solutions derived from suspensions of cercariae of *S. mansoni* incubated at 25°C. for 18 hours. To demonstrate this activity sodium hyaluronate must be included with the suspensions. Enzyme activity was uniformly present although found to vary considerably. The relationship between penetration of cercariae through the ground substance and enzyme activity will be discussed. A new viscometer designed specifically for viscosity ranges suitable for hyaluronidase assay was used for determinations in this study. Sodium salicylate given *per os* to mice over a seven day period failed to appreciably alter the number of adult worms recovered after exposure to known numbers of cercariae. Studies on *parenteral* administration of this salt in high concentration are now in progress.

Experimental production of acute pancreatitis. ROLF LIUM (by invitation) and STEPHEN MADDOCK. *From The Surgical Research Laboratory of the Boston City Hospital.* At the last meeting of this society a preliminary report on the production of acute pancreatitis was presented. Since that time the studies have been completed. A series of 100 cats, including controls, were subjected to ligation of both pancreatic ducts and the pancreas stimulated in a variety of ways: pilocarpine, esserine, acetylcholine and secretin with and without food. In the majority of above animals gross fat necrosis was found at the end of 24 to 48 hours. The amount of fat necrosis varied greatly, being most marked in animals which had been fed high fat meals prior to duct ligation. In control animals in which the

ducts were exposed by laparotomy no changes were observed at 48 hours whether the animals were starved, fed or chemically stimulated. The microscopic findings showed considerable variation in inflammatory reaction from edema with exudate and inflammatory cells to areas in which an entire lobule of the gland showed dissolution. Frank vascular lesions were not encountered but there were patchy areas of hemorrhage mingled with inflammatory reaction in a number of instances. Some sections showed damage to the walls of the blood vessels but in no instance was a gangrenous pancreatic necrosis observed. It is our opinion that this condition did not occur in cats because the pancreas is thin and lying free in the mesentery of the duodenum. The human pancreas in contrast with its thickness, dense capsule and peritoneal covering offers resistance to the extrusion of dammed up pancreatic juice.

The histogenesis of cells in pneumonia as seen in multiple lobe pneumococcus (type I) infections in dogs. CLAYTON G. LOOSLI. *Dept. of Medicine, University of Chicago, Chicago, Illinois.* A study was made of the origin of cells in the pneumonic exudate in the lungs of a single animal subjected to a pneumococcus (type I) infection in a different lobe at daily intervals until 5 lesions of increasing ages (4, 24, 48, 72, 96 hours) were produced. Twenty-two dogs weighing 8 to 12 kilograms were employed. The inocula (0.001 to 0.03 cc. of an 18-hour broth culture of type I pneumococcus in one cc. starch) were introduced into the lungs through a radiopaque catheter inserted into the bronchi via the tracheae with the aid of a fluoroscope. The extent and site of the infections produced by each inoculation was determined by x-rays. The animals were sacrificed with intravenous injections of pentobarbital sodium. The lungs were fixed intratracheally with Zenker-formol solution after the vessels were clamped to hold the blood in the capillaries, sectioned and stained by the hematoxylin-eosin-azure II method. By this technique, the pneumonic process was shown to be a dynamic one and the kinds of cells in the exudate varied with the age of the lesions. Polymorphonuclear leukocytes predominated in the 4 and 24 hour stages. During this time, however, large numbers of hematogenous lymphocytes and monocytes also entered the alveolar exudate. These cells hypertrophied and transformed themselves into macrophages which replaced the polymorphonuclear leukocytes. The local septal cells did not desquamate to form macrophages in the early stages but gradually enlarged and became most conspicuous on the alveolar walls in the 96-hour lesion when resolution began.

Hypoprothrombinemia with hemorrhage as a cause of death in the rat in hypervitaminosis A. CHARLOTTE L. MADDOCK (by invitation), S. B. WOLBACH and DOROTHY JENSEN (by invitation). *Department of Pathology, Harvard Medical School and Surgical Research Laboratory, Boston City Hospital.* During the course of an investigation on the bony changes resulting from excessive vitamin A administered to rats maintained on a rachitogenic diet for 3 weeks and over, an hemorrhagic diathesis developed in 90% of cases. Previous work in this laboratory on hypervitaminosis A, done on large series of animals maintained on normal diets, had never demonstrated striking hemorrhages. In this respect, the pathology resulting from excessive vitamin A was contrary to the reports of some other workers. A synthetic diet, devised by Bessey, was utilized. It was presumably complete in all respects except for absence of vitamin D, deficient P, and excess Ca. Vitamin K was not added because it is normally synthesized in rats' intestines. Growth on this diet was equal to that of stock animals for a period of 7 weeks. Subsequently weight gains declined somewhat. Rats, 41 to 71 days old, maintained on the diet since weaning, were given excessive doses of vitamin A for 5 days or longer. Multiple massive hemorrhages developed, most commonly located in the subdural space, epididymis, skeletal muscles and body cavities. Lymph nodes were hemorrhagic in appearance because of blood-filled sinuses. Hematocrit values fell as low as 13%. Prothrombin times were prolonged (70–183 seconds, controls 25 seconds or less). It seems that the Bessey diet plus hypervitaminosis A produces hemorrhages sufficient to cause death. If blood and tissue concentration of vitamin A can be brought to sufficiently high levels, fatal massive hemorrhages definitely related to hypoprothrombinemia will result.

Histochemical features of the renal basement membrane. J. F. A. MCMANUS (introduced by R. D. BAKER). *Department of Pathology, The Medical College of Alabama, Birmingham, Alabama.* The basement membrane (BM) of the kidney is colored with Schiffs reagent after periodic acid (PAS), a reaction originally described for mucin (Nature, Aug., 1946) which now appears to be characteristic of any carbohydrate. BM contains protein as demonstrable by digestion experiments. Fat shown with sudan IV or sudan black is absent normally but appears sometimes in disease. The BM is preserved in sections after almost any type of handling. The BM does not autolyze for a long time, suggesting the absence of any inherent enzyme synthesis. Accordingly, no phosphatase activity is shown in the BM of the human kidney at pH 4 to pH 9.5 with glycerophosphate, nucleic acid, adenylic acid, hexose diphosphate or lecithin as substrates except where the overlying epithelium is positive. The BM does not digest with diastase or hyaluronidase, not containing glycogen or hyaluronic acid in available form. In one case in which it was possible to study

the mitochondria of the tubule cells and the tubular BM, colloid droplets, replacing the mitochondria, were covered by thickened and abnormal BM. When the glomeruli are obsolescent or obsolete the corresponding tubules become atrophied and collapsed, with pigmentation of the epithelial cells in many instances. The BM becomes wrinkled and thickened, frequently duplicated and shows striking abnormality with PAS. These observations suggest a passive role for the BM, closely related to the activity of the tubular epithelium. It is likely that the BM is of the nature of chitin, as Lillie suggests; this is being tested with glycosidase.

The fate of aged leukocytosis-promoting factor of exudates. VALY MENKIN. *From the Agnes Barr Chase Foundation for Cancer Research, Temple University School of Medicine, Philadelphia, Pennsylvania.* In numerous earlier studies the writer has demonstrated in many inflammatory types of exudates a pseudoglobulin (cataphoretic studies show it to be an alpha globulin) which is capable of causing the specific growth of granulocytes in the bone marrow and to some extent of megakaryocytes. Besides this growth effect the substance is capable of inducing a discharge of immature polymorphonuclear leukocytes into the circulation, thus offering an explanation for the mechanism of leukocytosis with inflammation. This leukocytosis-promoting factor has been abbreviated as the LPF. The LPF can be preserved for several weeks *in vacuo* under phosphoric anhydride. After such an interval the factor tends to lose its biological potency. Gradually some chemical change occurs, presumably a spontaneous denaturation occurs. After several months the factor loses its initial solubility in an aqueous medium, and it generally becomes either very weak in its activity or else it becomes completely inactive. As shown previously, in such form the LPF can still be recovered as a soluble polypeptide which splits from the rest of the inactive molecule. The remaining portion of the inactive molecule of the original but now presumably denaturated LPF contains a leukopenic factor, and this explains the relative biological inactivity of the aged LPF. The leukopenic component counteracts the leukocytosis-promoting one. Both components can however be fairly well separated from each other by centrifugation and thus studied as individual entities.

Hypoplastic anemia induced in guinea pigs by 4-amino pteroyl glutamic acid. VIRGINIA MINNICH (by invitation) and CARL V. MOORE. *Dept. of Internal Medicine, Washington University School of Med., St. Louis, Mo.* Two folic acid inhibitors, a crude preparation and methyl folic acid, have been reported to produce anemia and leucopenia in chicks, mice, rats, and pigs. These changes have been prevented or reversed by folic acid. Of considerable interest, therefore, is the observation described in this report that another antagonist of folic acid, crystalline 4-amino pteroyl glutamic acid, induces similar changes in the peripheral blood of guinea pigs which cannot be prevented by either folic acid or liver extracts. Guinea pigs fed a stock ration were given daily by subcutaneous injection 0.5 to 5.0 mg. 4-amino pteroyl glutamic acid. They all lost weight and with few exceptions died in 11 to 28 days. Some of the animals developed a profound normocytic anemia and all a leucopenia (4000 to 750 WBC per cu. mm.) Half of the pigs had an agranulocytosis and half became thrombocytopenic. Varying degrees of marrow hypoplasia were found. If administration of the inhibitor was stopped after the hematologic changes had become profound, a few of the animals showed a striking reticulocytosis, rapid rise in all the cellular elements of the blood, and recovered completely. Highly purified liver extracts given with the inhibitor in doses of 4.5 to 5 USP units intramuscularly per day did not prevent the anemia or leucopenia. Folic acid in amounts 25 to 100 times that of the 4-amino pteroyl glutamic and injected simultaneously seemed to prevent the development of leucopenia or thrombocytopenia but not the anemia. Erythrocyte levels as low as 800,000 per cu. mm. were observed. Preliminary observations on other animals suggest that there may be considerable species difference both in the changes produced by this inhibitor and in the ability of folic acid to prevent them.

Purification of a substance from bovine serum which reduces the prothrombin time of aged plasma. F. L. MUNRO and MURIEL PLATT MUNRO (introduced by F. R. MILLER). *Charlotte Drake Cardeza Foundation, Department of Medicine, Jefferson Medical College and Hospital, Philadelphia, Pa.* Bovine serum contains relatively large amounts of a substance which reduces the prothrombin time (one stage method) of aged plasma. This substance has been named prothrombin A and "labile" factor by Quick (Am. J. Physiol. **150**, 405 (1947)). It is possibly the "factor V" described by Owren in "The Coagulation of Blood" Thesis, J. Chr. Gunderson, Oslo 1947, and the accelerating globulin described by Ware, Guest and Seegers (Science **106**, 41 (1947)). By adsorption on aluminum hydroxide followed by suitable elution in an alkaline medium, this substance may be obtained largely freed from other serum proteins. Since serum does not contain prothrombin or fibrinogen these substances are not present in the eluate as they are in similar eluates prepared from plasma. Calcium ions are essential for the stability of this substance in solution.

Adrenal cortical atrophy and liver damage

produced in dogs by feeding 2,2-bis-(parachlorophenyl)-1,1-dichloroethane (DDD). ARTHUR A. NELSON and GEOFFREY WOODARD (by invitation). *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington 25, D. C.* DDD is closely related to DDT and has considerable importance as an insecticide. Eleven dogs were fed DDD dissolved in corn oil in capsules at levels of 50 to 200 (usually 50 or 80) mg./kg./day for periods of 1 to 21 months. Six dogs died, 2 were sacrificed because of poor condition, and 1 was sacrificed while still in apparently good condition after 21 months. Two dogs survive, and will continue to be fed DDD. Gross and microscopic examination of the tissues of all 9 dogs sacrificed or found dead showed in every one a high degree of adrenal cortical atrophy. There was also high grade liver damage consisting chiefly of fatty degeneration and to a lesser extent of atrophy, necrosis and cirrhosis. In microscopic sections the adrenal cortical parenchyma was one-third to one-half its usual thickness, with much distortion of the normal architecture and alteration of the normal cellular appearances. The medulla appeared unaffected. Rats and a monkey fed DDD did not show the adrenal cortical atrophy, nor did dogs when fed the very closely related DDT. Of some dozens of compounds fed to 300 of our dogs, none except DDD has caused adrenal cortical atrophy even though several have caused severe liver damage; few have affected the adrenal in any way. The effect of DDD on the dog adrenal is a striking example of chemical specificity in the causation of organ damage.

Hereditary osteopetrosis of the rabbit. LOUISE PEARCE. *The Rockefeller Institute for Medical Research, Princeton, N. J.* An hereditary disease of the rabbit greatly resembling, if not identical with, the juvenile form of osteopetrosis of man has occurred in a rabbit breeding colony. The condition is inherited as an autosomal recessive character. It is present at birth and may be identified by the delayed and imperfect development of the teeth and by the characteristic x-ray appearance of the entire skeleton. The shadows are very dense and homogeneous with a lack of structural detail. Hydrocephalus is occasionally seen. Growth and development are retarded and beginning at about 2 weeks of age, progressive malnutrition with eventual cachexia develops. A hypochromic anemia which often reaches severe proportions is usually found. The condition is invariably lethal, the average survival being 4 to 5 weeks. The bones are hard but brittle; spontaneous fractures, however, are rare. In x-ray photographs of older cases, the ends of the ribs and of the long bones are frequently clubbed, the long bones and the femur in particular are shortened and the central portions of the bone shadows show translucent areas. In the younger cases, there is an excessive amount of abnormal spongy bone, the marrow cavities are markedly reduced and the marrow tissue is scanty. In older animals, the cavities are larger and contain more abundant marrow together with conspicuous amounts of abnormal fibrous tissue. Compared with normal litter mates, serum calcium was lower, serum phosphorus was lower in younger but higher in older cases and serum phosphatase was higher; the blood sugar content was lower and the blood cholesterol values were higher.

The influence of lipid ingestion of the thymol turbidity test. HANS POPPER, FREDERICK STEIGMANN, ALVIN DUBIN (by invitation) and HATTIE M. DYNIEWICZ (by invitation). *From the Hektoen Institute for Medical Research of the Cook County Hospital, Chicago, Illinois.* MacLagan's thymol turbidity test serves as a valuable indicator of liver cell damage A pathologic increase is less marked when liver cell damage is due to extrahepatic biliary obstruction. The turbidity depends upon relationship between lipo-proteins and globulins fractions. Extraction with ether but not with petrol-ether reduces markedly thymol turbidity in serum, oxalated and heparinized plasma. In 12 cases the turbidity in oxalated plasma and serum was identical but markedly reduced in heparinized plasma. Dicumarol had no effect. Adding heparin to 25 sera reduced turbidity proportionally to the amount added. These findings explained by an influence upon serum phospholipids prompted study of the effect of ingestion of various fats upon thymol turbidity. Usually 50 gm. butter produced an equal or higher rise than 4 egg yolks, 50 gm. cholesterol, 30 gm. lecithin or 30 cc. cod liver oil within 3 or 6 hours. It disappeared after 24 hours. The butter effect was markedly enhanced by simultaneous administration of choline (11 cases). Choline alone had a lesser influence upon thymol turbidity. Butter and choline raised the turbidity significantly in 23 hospital controls (average 2.6 units). In liver diseases (25 cases), especially in cirrhosis and obstructive jaundice, and in gastrointestinal disturbances (10 cases) the rise was much lower as expressed in units and even more so when expressed in percentage of the fasting turbidity. Further studies should elucidate the clinical value of the rise in thymol turbidity after butter and choline administration as simple test of fat absorption and the influence of choline upon intestinal absorption of fat.

Peroxidase in the lymphocytes of man in acute inflammation. JOHN W. REBUCK and ELIZABETH A. MONAGHAN (introduced by F. W. HARTMAN). *Dept. of Anatomy, Univ. of Minnesota, Minneapolis and Dept. Labs., Henry Ford Hospital, Detroit, Mich.* The cytology of the exudate in acute inflammation in man has been studied with an original technical procedure in which serial preparations

are obtained chronologically from a single lesion and are stained like blood smears. The epithelium is scraped away from a small area on the forearm. After inoculation with a suitable inflammatory excitant (egg-white) the lesion is covered by a cover-slip. Cells of the inflammatory exudate migrate to the under-surface of the cover-slip. When this has been accomplished (30 minutes to 1 hour) the cover-slip is removed, air-dried, and stained with Washburn's method for peroxidase. At the same time, another cover-slip is placed over the same lesion and the process is repeated at timed intervals throughout the cycle of inflammation. Transformation of the lymphocytes of man into macrophages proceeds in an orderly fashion in such a simple cycle of inflammation. It is accomplished by numerous gradual functional and structural changes in the lymphocytes as such. One of these changes is assumption of the positive peroxidase reaction by lymphocytic cytoplasm. By 9.5 hours, most of the lymphocytes in the area of inflammation contain numerous peroxidase-positive granules diffusely distributed throughout their cell bodies. The lymphocytes are actively phagocytic when this stage is reached. Sometimes it is apparent that a phagocytosed neutrophilic cytoplasmic bud within the lymphocytic cytoplasm has contributed a circumscribed area of peroxidase-positive granules to the cytoplasm of the lymphocyte (exogenous source). An occasional lymphocyte lacks such granules.

Effect of blood and oxygen on P. knowlesi infection in monkeys. R. H. RIGDON. *Department of Pathology, School of Medicine, University of Texas, Galveston, Texas.* Previous experimental studies with P. lophurae infection in ducks have shown that anoxemia is a significant factor during the course of the disease. Furthermore, life can be prolonged by placing these moribund birds in an oxygen chamber (Jour. Lab. and Clin. Med. **32:** 57, 1947). These observations were repeated with P. knowlesi infection in monkeys. A total of 27 Macacus rhesus animals was used. Temporary clinical improvement occurred when the infected monkeys were put into an oygen chamber with a concentration of 40–60% oxygen. Malarial infected monkeys when given intraperitoneal injections of human blood likewise showed clinical improvement. These observations would suggest that anoxemia is a significant factor in P. knowlesi infection in the monkeys. The destruction of the red cells by the plasmodia produces the anemic anoxia.

Arteriosclerotic lesions in pyridoxine deficient monkeys. JAMES F. RINEHART and LOUIS D. GREENBERG (by invitation). *Division of Pathology, University of California Medical School, San Francisco, California.* Tissues of 4 rhesus monkeys subjected to prolonged pyridoxine deficiency have been examined. The diet has been previously described (Fed. Proc. **5:** 222, 1946). Significant arteriosclerotic lesions have been found in 3 of the 4 animals. In 3 animals subjected to pyridoxine deficiency for 5 to 7 months 2 showed intimal fibrosis chiefly in the small arteries at the renal hilus. One animal subjected to pyridoxine deficiency for 14 months, showed also striking lesions in the coronary arteries. In this animal a broad layer of intimal edema and connective tissue proliferation was seen in major coronary arteries. This animal, in addition, presented an enlarged cirrhotic liver. These lesions may be related to the defect in tryptophane metabolism in the pyridoxine deficient monkey recorded elsewhere in this journal (Greenberg and Rinehart). The preliminary report is made because of its potential significance in the important problem of arteriosclerosis in man. Spontaneous arteriosclerotic lesions have not been encountered in the rhesus monkey and experimental production of arteriosclerosis in this species has not been recorded before.

The influence of folic acid on hemorrhage anemia in dogs. F. S. ROBSCHEIT-ROBBINS. *Department of Pathology, School of Medicine and Dentistry, University of Rochester, Rochester 7, New York.* The influence of folic acid on hemoglobin regeneration has been studied in experimental anemia due to blood removal in dogs. Folic acid was tested in daily doses of 5, 10, 20 and 40 mg. as supplements to lextron with and without iron. These materials were added to a basal ration of salmon bread and fed for a two week period. Net hemoglobin output was determined. Results vary. In some experiments a possible inhibition of hemoglobin production was evident when folic acid was administered. Other experiments demonstrate a negative effect.

Production in vitro from protein and other solutions of substances resembling "natural" agglutinins and precipitins. EDWARD C. ROSENOW. *From the Rare Metals Institute of the California Institute of Technology, Pasadena, California, and the Longview Hospital, Cincinnati, Ohio.* In experiments on the *in vitro* production of antibody with heat and H_2O_2, the yield from broth cultures and filtrates was greater than that from comparable suspensions andfi ltrates in NaCl solution, suggesting that constituents of the culture medium might be a source of antibody. The effect of keeping at room temperature for 24 hours and autoclaving for one hour protein and other solutions before and after adding 1.5% H_2O_2 and 10,000,000,000 pneumococci and streptococci $\overline{aa}$ per ml from dense suspensions in glycerol two parts and saturated NaCl solution one part was studied in parallel from the standpoint of antibody production. Agglutinins and precipitins for streptococci, pneumococci

and staphylococci were not formed at room temperature in 24 hours in the absence of H_2O_2 and either were not formed or were formed in minimal titer after adding H_2O_2 and 10,000,000,000 streptococci and pneumococci $\overline{aa}$ per ml to each of 5% solutions in NaCl solution of gelatin, peptone, potato starch, gum arabic, essential amino acids, 10% egg white and white and egg yolk $\overline{aa}$, dextrose brain broth, undiluted milk, and isotonic NaCl solution. Significant but variable titers were obtained from each of these solutions on autoclaving for one hour after adding 1.5% H_2O_2, and in all instances titers were far greater especially in the case of casein and amino acids on autoclaving after adding both H_2O_2 and the bacteria. The agglutinins produced in the protein and NaCl solution suspensions were filtrable, dialyzable and distillable before conjugation but were no longer dialyzable or distillable after conjugation onto normal globulin.

Preparation of radioactive gold colloids for use in the therapy of malignancies. GEORGE ROUSER (introduced by P. F. HAHN). *Vanderbilt University School of Med., Nashville, Tenn.* The use of radioactive gold colloids in the therapy of malignancies made a reliable method for producing them desirable. A procedure was devised in which the pH, rate and order of addition of reagents and temperature were controlled to give a stable colloidal gold solution of uniform particle size for various concentrations of gold. Colloids as concentrated as 5 mg./ml. were prepared with a uniform particle size using gelatine as a supporting colloid. The influence of pH, rate and order of addition of reagents and temperature is more pronounced as the concentration of gold is increased. The colloids were prepared by dissolving metallic gold in aqua regia, evaporation to dryness at 100°C under reduced pressure, solution of the auric chloride in water and its reduction to a metallic gold colloid with ascorbic acid, using gelatine as a supporting colloid. It was found that better control of particle size could be achieved by the slow addition of an aqueous solution of the auric chloride (chlorauric acid) to an aqueous solution of ascorbic acid and gelatine. The latter solution can then be heated to any desired temperature and the pH adjusted to any desired value by the addition of hydrochloric acid, sodium hydroxide or a suitable buffer without the danger of exposure to intense radiations.

Structure and repair of the olfactory mucosa in rhesus monkeys. EDWIN W. SCHULTZ. *Department of Bacteriology and Experimental Pathology, School of Medicine, Stanford University, Calif.* Observations were made on normal and zinc sulfate-treated mucosae. These were removed *in toto* and in a way which served to preserve their normal relationships to surrounding structures. They were fixed in Bouin's solution, embedded in paraffin and stained by a modification of the Bodian method of staining for neurofibrils. The normal mucosae were examined for variations in general structure, in the number and distribution of the olfactory cells, and for differences in the structure of the dendritic processes. Zinc sulfate solution constitutes a highly satisfactory agent for destroying the olfactory epithelium in regeneration studies. It induces a massive coagulation necrosis which is followed by a clean *en masse* separation of the necrotic epithelium from the underlying lamina propria. Stages of repair ranging from the 2nd day to the 8th month were observed. Interest centered primarily around the origin of the new epithelium, and the possible source of varying numbers of olfactory nerve cells present after the twelfth day.

Hydrolysis of DL-leucylglycylglycine by sera of tuberculous and normal rabbits. JULIUS SCHULTZ (by invitation) and CHARLES WEISS. *Laboratories of the Jewish Hospital, Philadelphia, 41, Pa.* The rate of hydrolysis of the tripeptide, DL-leucylglycylglycine (LGG), by aminopeptidase found in sera of tuberculous rabbits was compared with that of normal sera. Seven rabbits were injected intratracheally with 0.00095 mg. of a virulent Ravenel culture of tubercle bacilli. From 42 to 71 days later their sera were tested at least twice at intervals of about 10 days or more (in most cases). The average of 19 analyses was 0.41 ± 0.03% hydrolysis per minute at a concentration of 0.1 ml. serum per ml. test solution. The average rate for seven control rabbits, carried out simultaneously, was 0.184 ± 0.012% hydrolysis per minute at the same serum concentration. This represents more than a twofold increase in the rate of enzyme activity. These data are significant in the light of the work of Emmart and Seibert (J. Immunol. **50:** 143, 1945) who showed that the globulin fraction of tuberculous serum is tuberculostatic and of the finding of Fruton (J. Biol. Chem. **166:** 721, 1946) that this protein fraction contains the LGG-splitting peptidase. Since Weiss and Halliday (J. Immunol. **49:** 251, 1944) have previously observed that extracts of Cathepsin II, obtained from tissues more resistant to tuberculous infection, hydrolyses BAA faster than that obtained from less resistant organs, the present findings lend further support to the hypothesis that increased proteolytic activity is associated with increased resistance to infection in tuberculosis. The extent to which these results are peculiar to tuberculosis will be investigated.

Further studies on the hepatic potentiation of the estrogenic activity of triphenylchloroethylene. ALBERT SEGALOFF and RICHARD L. COPPEDGE (by invitation). *From the Departments of Medicine and Physiology, Tulane University and the Alton Ochsner Medical Foundation, New Orleans,*

Louisana. We have previously shown that less triphenylchloroethylene is required to produce vaginal estrus in spayed female rats when given intrasplenically than when given subcutaneously. This was interpreted as indicating that the liver increased the estrogenic activity of this compound. We have also demonstrated that more α-estradiol is required to produce vaginal estrus by intrasplenic injection than by subcutaneous injection. This was interpreted as indicating that α-estradiol was inactivated by the liver. When these studies with α-estradiol were repeated in partially hepatectomized animals, the amount required to produce vaginal estrus was greatly reduced, indicating further that the α-estradiol was inactivated by the liver. Accordingly triphenylchloroethylene was injected into partially hepatectomized female rats by the same two routes. There was increased potentiation of the estrogenic activity rather than the expected decrease. In the attempt to clarify this apparently anomalous result *in vitro* studies were undertaken. It was found that when an amount of triphenylchloroethylene below threshold for increasing the uterine weight of immature female rats was incubated with either liver slices or brei, then significant uterine weight increases were obtained. There is an optimal amount of brei which produces a greater potentiation than larger or smaller amounts of brei. Little or no potentiation of estrogenic activity is obtained with either very small or very large amounts of liver brei. It thus appears that there are two processes in the hepatic metabolism of triphenylchloroethylene: first the conversion to a more active compound, and then the inactivation of this product by oxidation or conjugation.

Chemotherapeutic effect of chloromycetin on experimental infection with psittacosis and lymphogranuloma venereum viruses. JOSEPH E. SMADEL and ELIZABETH B. JACKSON (by invitation). *Department of Virus and Rickettsial Diseases, Army Medical Department Research and Graduate School, Army Medical Center, Washington 12, D. C.* Chloromycetin, a new antibiotic, has previously been reported to possess chemotherapeutic activity against a number of experimental rickettsial infections. Growth in embryonated eggs of strains of psittacosis virus of pigeon or psittacine origin and of lymphogranuloma venereum virus is reduced by treatment with Chloromycetin. Administration of the drug 48 hours after the infection is almost as effective in prolonging the life of embryos as the prophylactic treatment. Doses of 0.0625 mg. per egg produce a distinct prolongation of life in infected eggs. Furthermore, the length of survival is directly proportional to the dose of drug over the range from 0.0625 mg. to 0.5 mg. per egg. Mice infected by the intraperitoneal route with the 6-BC strain of psittacosis survive 80 minimal lethal doses when given 0.75 mg. to 2.5 mg. of drug per day either by mouth or intraperitoneally. In addition, treatment is effective in mice when delayed for 6 days after infection, i.e., several days before death would be expected to occur. Mice infected by the intracerebral route with the P-4 strain of psittacosis and with lymphogranuloma venereum virus are benefited little if at all by treatment with Chloromycetin administered either by mouth or intraperitoneally. Data bearing on the explanation for the failure to obtain a chemotherapeutic effect following intracerebral infection with these viruses will be presented.

Experimental hypervolemia, time of onset and some associated physiological and chemical changes. HARRY SOBEL (by invitation) and JACOB FURTH. *Cornell University Medical College, New York City and Veterans Administration Hospital—Southwestern Medical College, Dallas 2, Texas.* Repeated blood volume determinations were made by the Evans Blue technique on mice bearing transplanted granulosa cell tumors. The blood volume began to rise after the tumors had reached a moderate size. Once this process was set in motion the blood volume increased rapidly and consistently from values below 10% of body weight to values as high as 28%, in the present series. This hypervolemia is not accompanied by an increase in the extracellular fluid (thiocyanate space). The plasma protein values may be normal and when they are reduced this is due to a decrease of plasma globulin concentration. The absolute amount of plasma albumin is, however, greatly increased and this increase roughly parallels the degree of hypervolemia. The blood urea levels may be high or normal in the presence of advanced hypervolemia. Other values will be given and the theory of hypervolemia discussed.

Production of arteriosclerosis in dogs with cholesterol and thiouracil. ALFRED STEINER, MARGARET BEVANS, and FORREST E. KENDALL (introduced by H. P. SMITH). *From the Research Service, First (Columbia) Division, Goldwater Memorial Hospital, Department of Hospitals, City of New York, and the Department of Medicine, College of Physicians and Surgeons, Columbia University.* The experimental production of intimal arteriosclerosis by the feeding of cholesterol to dogs whose thyroid function was depressed by thiouracil (Steiner and Kendall: Arch. Path. **42:** 433, 1946) has been repeated in two young dogs. The dogs, which were 4 months old at the beginning of the experiment, were maintained upon a diet of prepared dog food. 10 grams of cholesterol, dissolved in ether was added to the diet each day, the ether was evaporated off and the diet was moistened with milk before feeding. From 0.5 to 1.2 gms. of

thiouracil was administered each day. No additional fat was fed. This regimen resulted in serum cholesterol levels averaging above 1000 mg. per cent throughout the experiment. The animals were autopsied after 12 months. Extensive arteriosclerotic lesions developed in the aorta, in the coronary, mesenteric, renal and thyroid arteries and in the arteries of the upper and lower extremities. Of special interest were well defined lesions of the large cerebral arteries.

Desoxypyridoxine.—Morphologic and functional changes in acute pyridoxine deficiency. HERBERT C. STOERK (introduced by Dr. A. M. PAPPENHEIMER, SR.). *Dept. of Bacteriology, Harvard Medical School, Boston, Mass.* The administration of desoxypyridoxine to mice and rats produced marked atrophy of normal and of neoplastic lymphoid tissue and impairment of antibody responses. These changes failed to occur when pyridoxine-HCl was given together with the analogue. The severity of the manifestations of the disease, beginning from effective levels, was independent of the dosage of the pyridoxine antagonist. The loss of lymphoid tissue was not mediated by adrenal cortical hyperactivity, since adrenalectomy did not modify the extent of lymphoid atrophy in acute pyridoxine deficiency. Following the administration of desoxypyridoxine lymphosarcoma transplants showed marked regression associated with extensive pyknosis and caryorrhexis of tumor lymphocytes and apparent transformation of tumor cells into multinucleated giant cells. Rats treated with desoxypyridoxine showed marked impairment of immune responses. The anamnestic response, in mice and rats, was abolished in acute pyridoxine deficiency, whereas it was found unimpaired in adrenalectomized mice. The rate of disappearance of rat antibodies in rats (passive immunization) was not accelerated by the deficiency. Tyzzer's disease occurred in many instances in mice treated with desoxypyridoxine. This infection had never before been observed in the strain of mice employed.

Some factors associated with variations in nitrogen and ash content of bone. LOUIS J. STROBINO (introduced by LEE E. FARR). *Alfred I. du Pont Institute of the Nemours Foundation, Wilmington, Delaware.* Reports in the literature indicate considerable variation in nitrogen content of bone. The present study sought to determine some of the factors responsible in beef and human bones. Samples from the interepiphyseal cortex of long bones carefully cleaned of extraneous material were dried to constant weight and analyzed for nitrogen and ash. Nitrogen was determined by the gasometric micro-Kjeldahl method of Van Slyke and ash was estimated gravimetrically after heating in a muffle furnace at 600°C. Serial samples were obtained longitudinally between the epiphyses and circumferentially at various levels of the bone shaft. In addition, subperiosteal bone samples were compared with adjacent samples contiguous to the endosteum. In young subject's bones, an area of high ash, low nitrogen content was observed with a regular increase in nitrogen content and corresponding decrease in ash content as the epiphyses were approached. The position in the shaft of the area of maximum ash-nitrogen ratio was found to be characteristic of the bone. With increasing age, longitudinal variations in ash-nitrogen ratio of the shaft gradually diminished so that by the sixth decade of life, the ash-nitrogen ratio was approximately constant and was somewhat greater than the maximum ratio found in bones of young subjects. Analyses of beef bone were found to correspond to those of humans. When the subject's age and the geometric position of the samples are considered, there becomes apparent a reasonable constancy in nitrogen and ash content of different bones.

Prolonged parenteral plasma produces hyperproteinemia and proteinuria in dogs and maintains nitrogen equilibrium and health. ROGER TERRY (introduced by G. H. WHIPPLE). *School of Medicine and Dentistry, University of Rochester, Department of Pathology.* In seven separate experiments five normal dogs received homologous heparinized plasma parenterally as a sole source of nitrogen for periods of 15–92 days. If the period of plasma administration is preceded by a fairly long period of protein starvation, there is a strong positive nitrogen balance for the first 7–10 days of plasma injections and nitrogen equilibrium is maintained thereafter. No excessive excretion of nitrogen is noted in the after-period. If adequate calories and vitamins are supplied, weight balance is maintained and the injected plasma seems to be somewhat better utilized. Blood plasma concentrations rise rapidly and vary from 9.5–11.5 gms. % during the period of plasma administration. Soon after the hyperproteinemia is noted, proteinuria occurs in measureable amounts. All protein fractions except fibrinogen are present in the bladder urine and are apparently excreted at a constant rate. Globulins form about 30% of the urinary protein. After plasma injections are stopped and after the blood plasma concentration falls below 9 gms. %, the proteinuria rapidly diminishes and ceases entirely, indicating no permanent kidney damage. These experiments confirm the concept of a "dynamic equilibrium" between the plasma proteins and tissue proteins. (One dog was maintained in health and nitrogen equilibrium for 3 months with plasma as a sole source of nitrogen.) They also suggest (1) a "ceiling" of utilization of plasma protein, and (2) a renal threshold for protein molecules, in that proteinuria occurs when the circulating plasma

concentration is maintained at levels greater than 9–10 gms. %.

Specificity of human serum antihyaluronidase for antagonism of a particular species of bacterial hyaluronidase. ROBERT T. THOMPSON and FRANCES E. MOSES (introduced by M. A. BLANKENHORN). *Departments of Internal Medicine and Biochemistry, College of Medicine, Univeristy of Cincinnati.* McClean's demonstration of the antigenicity of hyaluronidases in rabbits (1943) indicates that an antihyaluronidase serum is specifically antagonistic to the particular species of bacterial hyaluronidase which was used as the antigen. The present study investigates this principle in the sera of humans who suffered bacteremia due to various bacteria. A modification of the mucoprotein clot prevention (M.C.P.) test (McClean) was used to study the sera of eight patients. Serial sera of three patients with pneumococcus bacteremia, one with staphylococcus bacteremia, one with B. coli Communis bacteremia, and one with B. typhosus bacteremia were titrated for antihyaluronidase against each of three hyaluronidases from culture filtrates of pneumococcus, staphylococcus, and Cl. welchii. Sera of the three patients with pneumococcus bacteremia exhibited eight-fold increases of antagonism to pneumococcus hyaluronidase, but no increase of antagonism to staphylococcus and Cl. welchii hyaluronidases. Sera of the patient with staphylococcus bacteremia exhibited an eight-fold increase of antagonism to staphylococcus hyaluronidase, but no significant increase of antagonism to pneumococcus and Cl. welchii hyaluronidases. Sera of the patients with B. coli and B. typhosus bacteremia exhibited no increase of antagonism to any of the three hyaluronidases. Serial sera of two other patients with pneumococcus bacteremia and empyema exhibited increases of antagonism to pneumococcus hyaluronidase, but no antagonism to Cl. welchii hyaluronidase. It has been reported previously that this human serum antagonism to pneumococcus hyaluronidase is not specific for type of pneumococcus.

Further studies on a fat-soluble biotin-active material (FSF) from plasma. WILLIAM TRAGER. *Department of Animal and Plant Pathology, The Rockefeller Institute for Medical Research, Princeton, N. J.* Experiments in addition to those previously reported have again shown that the oil obtained from hydrolyzed horse plasma has biotin-like activity for chickens as well as for bacteria. When chickens fed a diet high in egg-white were injected intramuscularly with this oil in the maximum dosage which could be well tolerated (equivalent by microbiological assay with *Lactobacillus casei* to 1 microgram of biotin per week), they developed less dermatitis of the mouth and feet than did uninjected chickens on the same diet. The effect was almost as good as that obtained with the injection of 1 microgram of biotin itself. Chickens similarly injected with oleic acid showed as severe a dermatitis as the untreated controls. Hence, although oleic acid has about 3 times the biotin activity for *L. casei* of the oil from horse plasma, it cannot account for the biotin-like effect of the latter material in chickens. Fractionation of the oil from horse plasma has yielded preparations which are probably not pure but which on a weight basis have as high a biotin activity for *L. casei* as oleic acid. These preparations are solid, whitish, waxy materials at room temperatures. Their activity in chickens has not yet been tested. FSF activity (the biotin activity liberated by acid autoclaving and removed by ether from alkaline suspension) is much higher in the plasma of egg-laying chickens and ducks than in the plasma of males or non-egg-laying female birds of comparable age.

AMERICAN INSTITUTE OF NUTRITION

TWELTH ANNUAL MEETING

Atlantic City, New Jersey, March 15, 16, 17, 18, 19, 1948

(For possible corrections in any of the following abstracts see the next issue.)

Studies on reproduction and lactation in rats and mice maintained on synthetic diets. SISTER ANN MIRIAM ALLGEIER (by invitation), ALBERT J. SICA (by invitation), LEONORA MIRONE (by invitation), FRANK P. PANZARELLA (by invitation), and LEOPOLD R. CERECEDO. *Department of Biochemistry, Fordham University, New York.* Experiments on reproduction and lactation in rats and mice maintained on synthetic diets have shown the following facts: 1. There is a close connection between birth-weight in the rat, nutrition of the mother before and during pregnancy in the rat and the mouse, and survival of the young. 2. The first four days in the life of the mouse and the first three days in the life of the rat are critical periods. Non-survival during these periods points to inadequacy for reproduction of the diet received by the mother. Such failures fall into the same category as still-births, resorptions and abortions. 3. In all studies on reproduction and lactation, the previous nutritional history of the animals must be considered. Our experiments with rats have shown that pteroylglutamic acid, given only during pregnancy and lactation, has some beneficial effect. However, a more pronounced effect on both reproduction and lactation was obtained when the supplement was given from weaning. The relative merits of so-called "longterm" and "short-term" experiments in the study of the nutritional requirements for reproduction and lactation will be discussed.

Effect of arsonic acid derivatives in stimulating growth of chicks fed certain diets. H. R. BIRD, A. C. GROSCHKE (by invitation), and MAX RUBIN (by invitation). The growth of chickens fed diets high in soybean meal and deficient in the unknown dietary factory found in fish meal and in cow manure was improved by the addition to the diet of 0.005% of 3-nitro-4-hydroxyphenyl arsonic acid. The effect of higher or lower levels was less favorable. This compound did not function as a substitute either for the unknown factor or for methionine. On the contrary, with diets containing raw soybeans, the arsonic acid derivative, the unknown factor, and methionine were mutually supplementary. Such a supplementary effect could not be demonstrated with diets containing commercially heated soybean meal because addition of the unknown factor induced essentially optimal growth in the absence of other supplements. The arsonic acid was effective when fed with 35% or with 70% of commercially heated soybean meal, but less effective than the unknown factor. Other compounds comparable in their activity to 3-nitro-4-hydroxyphenyl arsonic acid were phenyl arsonic acid, p-hydroxyphenyl arsonic acid, and m-nitrophenyl arsonic acid. The following were tested and found inactive: p-chlorophenyl arsonic acid, sodium arsenate, m-nitrobenzensulfonic acid, salicylic acid, 3-nitrosalicylic acid, and sodium p-phenolsulfonate.

Protein requirements of ten women and adequacy of estimated quantities for ten weeks. MILDRED L. BRICKER (by invitation), RUTH F. SHIVELY (by invitation), and JANICE M. SMITH. *Agricultural Experiment Station, Dept. of Home Economics, University of Illinois, Urbana, Illinois.* Ten girls, 19–30 years of age, cooperated in this study of nitrogen balances which lasted for 105 days. Four intakes of nitrogen (approximately 1.8, 3.1, 0.2, and 4.3–6.7 grams daily) were ingested during four periods. The first two periods of ten days each and the third low nitrogen period of 15 days duration were used in estimating the minimum amount of a protein mixture required for nitrogen equilibrium. During the fourth period, 70 days in length, the adequacy of the calculated protein requirement was tested for each of the subjects by continuous nitrogen balance data, weekly hemoglobin determinations, red cell counts and performance tests. The protein mixture, composed of bread; rolled oats; lean beef; light cream and white potatoes in proportions to furnish 59.5; 10.5; 14.7; 13.4 and 1.9% of the protein intake respectively, had an average nutritive index (biological value x true digestibility) of 60.9. The quantities of protein of this composition needed for nitrogen equilibrium plus the amounts calculated to be needed for "adult growth" were found to be 31.7, 35.2, 27.1, 30.2, 30.9, 26.9, 29.4, 42.2 and 35.2 grams daily for each of nine subjects. Throughout the ten-week balance period, nine of the subjects remained in definite positive nitrogen balance; hemoglobin and red cell values were maintained at initial levels and performance tests including critical fusion frequency of flicker, speed of tapping, endurance on a bicycle ergometer and mental and emotional responses showed no deterioration of function.

Niacin and protein relationships in corn diets for growing pigs. W. T. BURNETT (by invitation), R. C. MILLER, and R. A. DUTCHER. *Pennsylvania State College, State College, Pennsylvania.* Young growing pigs were fed high corn diets containing additional protein from either casein or corn gluten.

Diets containing 15 and 20 per cent of protein were compared. In addition all of the pigs received thiamine, riboflavin, pyridoxine, pantothenic acid, inositol, p-aminobenzoic acid, carotene, vitamin D and a complete salt mixture. Characteristic symptoms of niacin deficiency did not occur with pigs fed combinations of corn and corn gluten at either the 15 or the 20 per cent levels of protein intake, or with pigs fed the 20 per cent protein diet containing 5 per cent of casein. Pigs on the 15 per cent protein diet, containing 5 per cent of casein, grew more slowly unless given supplemental niacin. Niacin was without any apparent effect on growth on the exclusive corn diets at either protein level. The N′-methylnicotinamide excretion indicated increased methylation with the niacin supplemented pigs, except in the case of those pigs receiving the 20 per cent protein diet containing 5 per cent of casein. Further increased methylation occurred when both choline and niacin were added to the diet simultaneously. However, the feeding of niacin and choline in combination resulted in an unfavorable growth response. There was little if any correlation between N′-methylnicotinamide excretion and growth response in these studies.

Achromotrichia produced in mice on a cooked egg diet. EDITH M. CARLISLE (by invitation) and HELEN T. PARSONS. *Dept. of Home Economics, University of Wisconsin, Madison.* The hair of black mice of a non-cancerous strain, C_{57}, turned an all-over silver gray on a diet of egg white and yolk cooked with salts and later cerelose added. These mice developed low hemoglobin levels and showed marked edema. Another group of mice were fed a similar ration except the yolks and whites were cooked separately and the mineral salts added later. These mice showed little or no change in the color of their original coat of hair and maintained normal hemoglobin levels. The addition of copper to the ration of the strikingly gray mice stimulated a rapid growth of thick black hair clearly observed in areas of skin after depilation. Their hemoglobin values rose rapidly. Hence the cause of the grayness was attributable to a relative unavailability of the copper of the ration. It has been shown in other laboratories that a deficiency of copper produces in the rabbit an achromotrichia which seems to appear before the anemia; and also that the copper of egg yolk is relatively unavailable to the rat. These present experiments show that copper undergoes a change when cooked with egg which renders this mineral unavailable to the mouse.

Changes in the water content of the albino rat during the first week of dietary rehabilitation. ESTHER DACOSTA (introduced by R. E. JOHNSON) and RUTH CLAYTON (introduced by R. E. JOHNSON). With the technical assistance of Corporal GARY G. LALANNE. *U. S. Army Medical Nutrition Laboratory, Chicago 9, Illinois.* After 13 weeks of dietary restriction, i.e., (a) low calorie, (b) with 1% protein from carrots, and (c) with 10% special salt and 5% protein, 8 rats from each of these groups and controls were sacrificed and the water content of the total carcasses determined. Twenty-four animals from each group were rehabilitated on diets isocaloric with the control diet but in experiment one containing 60% protein, and in experiment two, 35% fat. In experiment one the water content of the carcasses of the controls was 64%. The water content of the carcasses of the refed low calorie rats was 68.8% after 24 hrs., 65.7% after 48 hrs., 65.5% after 72 hrs. and 67% after 96 hrs. The refed carrot diet rats showed 68.6%, 68.5%, 68.1% and 65.7% water content after 24, 48, 72 and 96 hrs. of refeeding. The refed high salt animals had a water content of 68.8%, 69.3%, 67.7% and 66.5% in the same units of time. During the 13 preliminary weeks, the controls gained 32%, the restricted animals lost between 21 and 28%. In one week of rehabilitation, the refed animal gained from 26 to 37%. In experiment two the changes in water content and weight during restriction were of the same order as in experiment one but during rehabilitation water was lost more rapidly and weight gained more rapidly than in experiment one.

Thus in one week of feeding a high protein diet after restriction there was a gradual dehydration, and on a high fat diet a rapid dehydration.

Studies on the cellulose growth factor for chicks. FRANK DAVIS (by invitation) and GEORGE M. BRIGGS. *Department of Poultry Husbandry, University of Maryland, College Park, Maryland.* Davis and Briggs (J. Nutrition **34:** 293, 1947) reported the growth-promoting action of cellulose in purified chick diets. Using the same experimental procedure and basal ration, degradation products and substances associated with and structurely similar to cellulose were tested for their effect on chick growth. Mixed sawdust composed of white and yellow pine, levulenic acid and d-xylose were fed separately to day-old New Hampshire chicks at various supplemental levels at the expense of glucose in the ration. Statistical analysis of chick weights at the end of repeated 4-week trials showed that the growth increments obtained with 0.1% of levulenic acid and 10% of mixed sawdust were highly significant. Supplementation with 0.05 through 2% of d-xylose gave no appreciable growth response. Preliminary experiments with furfuramide and d+cellobiose indicate that these compounds possess little or no cellulosic growth activity. Furfural, however, at the 0.1 and 0.3% supplemental levels gave a growth response similar to 0.1% of levulenic acid. These observations indicate that 10% of mixed sawdust, 0.1% of levulenic acid, and 0.1 through 0.3% of furfural ca

probably be substituted for 5% of cellulose in chick purified diets with equal growth response.

Pteroylglutamic acid balance studies on monkeys. PAUL L. DAY, DOROTHY S. GAINES (by invitation), MARION MCKEE (by invitation), PHYLLIS SCROGGIN (by invitation), and RAYMOND HOUCHINS (by invitation). *Department of Biochemistry, School of Medicine, University of Arkansas, Little Rock.* Juvenile rhesus monkeys were given a diet known to produce the anemia and leucopenia of pteroylglutamic acid deficiency, supplemented with the amounts of pteroylglutamic acid (PGA) or related materials as indicated below, for periods of 21 days or more. Daily urine and feces collections were made. Assays for free PGA were made on the urine, and for free and conjugated PGA on the feces, using *Streptococcus faecalis* and a suitable conjugase. When the diet was supplemented with 100 micrograms of PGA daily, the average daily output in micrograms was as follows: urine, 4.9; feces, 5.8; total, 10.7. When the daily dietary supplement of PGA was increased to 1000 micrograms the output in micrograms was: urine, 21.0; feces, 9.1; total 30.1. In another experiment the following supplements were made to the diets of comparable monkeys: 2.5 grams Difco yeast extract containing approximately 100 micrograms of PGA (largely as conjugase); 100 micrograms of PGA; 100 micrograms of pteroic acid; negative control receiving no supplement. The outputs of PGA in urine and feces of these animals were essentially the same. In summary, a ten-fold increase in PGA intake resulted in a four-fold increase in urinary output, but did not appreciably alter the fecal output of the vitamin. Monkeys receiving microbiologically equivalent amounts of PGA, PGA conjugate (as yeast extract), or pteroic acid, and a negative control, all excreted equivalent amounts of microbiologically active material in the urine and in the feces.

Changes in dehydro and total ascorbic acid of cantaloupes on standing. KATHERINE J. ELLIOTT (by invitation), SHIH DZUNG CHEN, (by invitation) and CECILIA SCHUCK. *Purdue University.* In the past the original ascorbic content of foods and losses on standing have usually been reported in terms of reduced ascorbic acid only. It is now questionable as to whether reported losses of ascorbic, when only the reduced form has been determined, represent true losses. Recent studies have shown that decreases in the reduced ascorbic acid of cabbage could be accounted for largely by conversion to dehydro-ascorbic acid. (Science **103**, 196 (1946); Journal American Dietetic Association **23**, 223 (1947). The present study was carried out to determine the amount of dehydro, reduced and total ascorbic acid in cantaloupes immediately after cutting and after standing in the refrigerator and at room temperature for 24 hours. The melons were cut crosswise through the center with a plastic knife and a circle removed for analysis. Half of the remainder of each melon was then placed in the refrigerator and half allowed to stand at room temperature. Both halves were wrapped in waxed paper. The diphenylhydrazine procedure of Roe and Oesterling, (Journal Biological Chemistry 152, 511, (1944), was used to determine the dehydro and total ascorbic acid and the reduced ascorbic acid was obtained by difference. The findings indicate that there was partial conversion of reduced ascorbic acid to the dehydro form both in the refrigerated melons and those held at room temperature, the change being somewhat greater in those held at room temperature. The results suggest a need for the determination of total and dehydro ascorbic acid in other foods under different conditions of handling and preparation.

Influence of liver extracts on a sulfathiazole induced dietary deficiency in rats. GLADYS A. EMERSON and CHARLES W. MUSHETT (by invitation). *Merck Institute for Therapeutic Research, Rahway, N. J.* A number of sulfonamides have been fed to rats as adjuncts to purified diets containing all identified vitamins except pteroylglutamic acid. A deficiency state evidenced by a failure in growth and a dyscrasia was noted after 4–5 weeks on test. Animals maintained on the ration containing 1% sulfathiazole made a rapid response to the administration of certain (4 out of 5) parenteral liver extracts as did those receiving adequate dosage of pteroylglutamic acid. Since the content of pteroylglutamic acid or its conjugates in the liver preparations was insufficient to account for the curative action, the presence of an unknown factor is indicated. The parenteral extract which was without effect in these tests was active in the treatment of the deficiency state induced by the feeding of a low protein diet (Daft and Sebrell). A limited number of experiments have been conducted with mice maintained on rations containing 1.25% sulfathiazole. This species also responded to a liver extract.

Anemia in chronic choline deficiency in the rat. R. W. ENGEL (introduced by W. D. SALMON). *Laby. of Animal Nutrition, Alabama Polytechnic Inst., Auburn.* Weanling rats of the Alabama Agricultural Experiment Station (AES) strain developed an anemia in 2 to 3 months when fed diets deficient in choline or substitute nutrients. The anemia was characterized by a reduction in hemoglobin to as low as 5 gm. per 100 ml blood, a reduction in total red blood cells to as low as 3.5 million per cmm., and a lowered color index. The severity of this condition was directly related to the degree of deficiency of the diet in choline or substitute nutrients. The diets employed ranged in composition from 4.5% to 18% in extracted casein; from 20% to 40% in degerminated corn

grits or alcohol-extracted peanut meal; and from 2 to 20% in lard. All diets contained 4% salt mixture and sucrose to make 100%. The diets were fortified with adequate amounts of thiamine, riboflavin, pyridoxin, niacin, inositol, ca pantothenate, a-tocopherol, and vitamins A and D in the form of cod liver oil. The anemia was prevented by fortifying the diets with choline Cl (2 gm./kg.) or with DL-methionine (3 gm./kg.). The sodium salt of pteroyl di glutamic acid (1 mg./kg.) was ineffective as a curative agent when added to a choline-deficient diet.

Occurrence of B-vitamins in tissues of rats fed rations satisfactory and unsatisfactory for reproduction. GLADYS EVERSON (by invitation), ELEANOR WILLIAMS (by invitation), ELIZABETH WHEELER (by invitation), PEARL SWANSON, MARGARET EPPRIGHT (by invitation) and MATTIE SPIVEY (by invitation). *The Nutrition Laboratory, The Foods and Nutrition Section, Iowa Agricultural Experiment Station, Iowa State College, Ames.* Toxemia of pregnancy in addition to other manifestations of unsatisfactory reproduction, i.e., sterility, resorptions and production of non-viable young have been encountered in female rats receiving a ration containing partially dried autoclaved pork muscle and five parts of yeast as the main sources of protein and the B-vitamins. In an effort to determine the dietary cause of these unsuccessful reproductions, assays have been made of the respective concentrations of thiamine, riboflavin, nicotinic acid, biotin, pantothenic acid, and folic acid in the liver, carcass, and fetuses of pregnant rats fed the ration. For comparison, similar assays have been conducted on tissues of animals receiving a modified Steenbock stock ration adequate for the support of normal reproduction. Vitamin stores were studied throughout two pregnancies. Analyses showed that the two rations provided widely differing amounts of pantothenic acid and biotin, the pork-containing diet being less rich in these factors than the stock ration. The hepatic tissues of rats ingesting the lower intakes contained smaller quantities of both biotin and pantothenic acid than did those of the control animals. The concentration of the two vitamins in the fetal tissues of the pork-fed rats also was less than that in the young of females successfully undergoing reproduction. The occurrence of all six B-vitamins in hepatic, carcass, and fetal tissue will be given for animals receiving both types of rations.

The pantothenate requirements of the mouse, with observations on the role of biotin, inositol, and p-aminobenzoic acid. PAUL F. FENTON and GEORGE R. COWGILL. *Yale Nutrition Laboratory, Department of Physiological Chemistry, Yale University.* Male mice of the C57, C3H and A strains were placed on a pantothenate deficient basal diet for one week. After this depletion they were fed the identical basal diet with graded doses of pantothenic acid. At all levels of pantothenate studied the C57 strain mice grew better than those of the C3H and A strains. Deficiency symptoms (achromotrichia, alopecia, dry dermatitis) appeared early in the C57 mice on low intakes of pantothenate. C3H and A strain mice, on the other hand, showed only occasional slight alopecia. The blood picture (red and white cell counts and hemoglobin levels) of C3H and A strain mice remained essentially within normal limits. C57 mice on low pantothenate diets showed a lowering of the red cell count and hemoglobin levels, while occasional animals showed abnormally high white cell counts. The pantothenate levels in muscle and liver were determined microbiologically. Biotin injections were found to speed the restoration of hair and the return of normal color. A large group of C57, A and I strain mice were placed on one of four synthetic rations: a) complete, b) deficient in inositol, c) lacking p-aminobenzoic acid, and d) without inositol or p-aminobenzoic acid. No clear-cut symptoms of inositol or p-aminobenzoic acid deficiency could be demonstrated. Another group was fed these same diets after being depleted of pantothenic acid. A fourth group received a diet moderately low in pantothenate and devoid of p-aminobenzoic acid and inositol. Studies of the cecal flora were made.

Thiouracil content of poultry tissue following prolonged feeding of the compound. A. L. FRANKLIN (by invitation), J. W. BOEHNE III (by invitation) and T. H. JUKES. *Lederle Laboratories Division, American Cyanamid Company, Pearl River, N. Y.* Numerous studies indicate that the feeding of thiouracil to poultry improves carcass grade and increases feeding efficiency. Information is lacking, however, regarding the extent to which thiouracil is stored in the tissues, and information regarding this point is necessary before thiouracil may be fed to chickens which are intended for human consumption. Satisfactory chemical methods for the determination of thiouracil in tissues are not available. A biochemical method was used in which the tissue thiouracil content was ascertained by determining the degree of depression of thyroid function in rats which had been fed the dried tissue for a three week period at a 50% level in the diet. This was accomplished by measuring the accumulation of an injected dose of radioactive iodine and the formation of thyroid hormone from this iodine by the thyroid gland. After feeding thiouracil at a 0.2% level to chickens for 3 weeks the thiouracil content of the muscular tissue was found to be 3 ±2 mg. %. When the thiouracil supplementation was discontinued for 24 and 48 hours the tissue thiouracil content fell to about 0.1 mg. % and less than 0.05 mg. % respectively. Cooking the meat for one hour at 130° had no effect on the level of thiouracil; however storage of the

tissue at 4° for one week resulted in the disappearance of most of the compound from the tissue.

The utilization of nicotinic acid by pregnant women. ERNESTINE I. FRAZIER (by invitation), THELMA PORTER and MARY JANE HUMPHREY (by invitation). *Dept. of Home Economics, Univ. of Chicago.* The intake of nicotinic acid and the urinary excretion of its acid hydrolyzable products and N^1-methylnicotinamide of 7 primaparas on self-selected diets was studied for 8 day periods during each of the last 5 months of pregnancy. The average daily intakes of 6 of the primaparas ranged from 12.8 to 20 milligrams. The seventh subject had an average intake of 33.7 milligrams per day including 20 milligrams of nicotinamide. On intakes up to 20 milligrams there was an increase in the excretion of N^1-methylnicotinamide during the last trimester of pregnancy. The level of excretion of the subject on the higher intake remained quite constant through the eighth month after which there was a decrease in excretion of the methyl derivative. The average daily excretion of N^1-methylnicotinamide for all subjects ranged from 13.0 to 16.2 milligrams from the fifth through the ninth month of pregnancy. The level of excretion of the acid hydrolyzable fraction showed only slight variations with a range of average daily excretion of 0.76 milligrams on an average intake of 12.8 to 1.57 milligrams on the average intake of 33.7. On an average intake of 12.2 milligrams of nicotinic acid per day the average daily excertion of N^1-methylnicotinamide for 44 clinic patients was 13.3 milligrams. When grouped according to months of pregnancy, the same general trend in increased excretion was noted. There was considerable variation in percentage return of test dose as N^1-methylnicotinamide, irrespective of daily intake, with a trend toward increased return with increased duration of pregnancy.

The absorption of iron from beef by women. RUTH FRENCHMAN, EUPHEMIA D. BOROUGHS, FRANCES A. JOHNSON (introduced by HAZEL M. HAUCK). *N. Y. State College of Home Economics, Cornell University, Ithaca, N. Y.* Five young women 18 years of age and in good health, with normal serum iron, hemoglobin and red cell counts, were placed on a diet containing 7 mg of iron which was judged to be the lowest intake that would be adequate. After six weeks on the basal diet, two 100 gm. patties of ground beef ($\frac{3}{4}$ lean meat, $\frac{1}{4}$ fat) containing 3.4 mg. of iron were added, one at breakfast and one at lunch. The increase in retention after the addition of the beef was considered as iron absorbed from the beef. The absorption of iron from beef for four out of five subjects were 32, 45, 58 and 57 per cent. This led to the conclusion that young women 18 years of age on an adequate diet absorb from about one-third to more than one-half of the iron of beef. Another interpretation of the results may be the correct one: that the beef improved the absorption of the iron of the entire food mixture, rather than itself contributing all of the iron absorbed after it was added to the diet. If the amount of iron added to the food residue as it passes through the gastro-intestinal tract is negligible, the mean absorption from the basal diet was 11 per cent and from the basal diet plus beef was 21 per cent.

The turbidimetric determination of infused fat in blood after intravenous administration of fat emulsions. ROBERT P. GEYER (by invitation), GEORGE V. MANN (by invitation), and FREDRICK J. STARE. *Dept. of Nutrition, Harvard School of Public Health and Dept. of Biological Chemistry, Harvard Medical School, Boston.* A turbidimetric method for the determination of the rate of disappearance of infused fat from the blood has been developed. After the intravenous administration of a fat emulsion, 20 cmm. samples of blood are withdrawn at intervals from the animal and are added to 5% dextrose solution. Following centrifugation at slow speed, the supernatant is decanted and to it are added concentrated ammonium hydroxide (to remove extraneous turbidity) and hydrogen peroxide (to remove hemoglobin). The turbidity which remains after heating to 60°C. is determined by means of a photoelectric colorimeter using a 420 mμ filter. Since the optical density is a linear function of the concentration, the latter can be obtained from a standard curve. Results obtained by the use of this method agree well with those obtained by the use of either the micro-oxidative or the chylomicrograph technique. The term "fat tolerance curve" is used to designate the curve which results when the concentration of fat in the blood is plotted against time. Such curves have been determined for the rat, dog, and rabbit after the administration of an emulsion containing 30% coconut oil and 3% phosphatides. The first two species remove the fat at rapid rates, while the rabbit does so more slowly. When mineral oil was substituted for the coconut oil in the case of the rabbit, the oil was removed from the blood at a very slow rate.

Minimal protein requirement for growth in the rat. MARIANNE GOETTSCH. *Department of Chemistry, School of Tropical Medicine, San Juan, Puerto Rico.* The growth and reproduction of rats was observed on diets ranging from 7.1 to 19.1 per cent protein, supplied by polished rice, 56.4 perts; red kidney beans, 28.2; and casein, 15.4. Minerals, vitamins and cornstarch were added. The diets contained 1.5 per cent fat and 3.4 Calories (calculated) per g. The "true digestibility" and biological value of the N were determined by appropriate metabolism tests on rats to be respectively 84.1 and 74.7. Diets containing 16.7 or greater per cent of protein supported growth in several genera-

tions of rats that was equal to that of controls on an adequate diet. Rats receiving 14.3 per cent protein from the 21st day, grew approximately as well as controls, but in successive generations, they grew less rapidly. With further reduction in dietary protein, the rate of growth decreased in rats given the diets from the 21st day. Upon 7.1 per cent protein there appeared the hepatic necrosis associated with methionine deficiency. The maximum value for the coefficient of protein utilization for growth, as determined by Osborne and Mendel, occurred on diets containing 14.3, 11.9, and 9.5 per cent protein and was 2.30. This is further proof that the 14.3 per cent level was inadequate. The 16.7 per cent diet contained 7.8 mg. T. N. per Calorie. Experiments are in progress to determine whether: (1). Approximately 5.0 mg. net T. N. per Calorie of any protein is the minimum for growth. (2). The coefficient of utilization of the most efficient protein, under these conditions, is approximately 3.70.

Urinary excretion of B vitamins in persons on normal and restricted diets. GRACE A. GOLDSMITH and HERBERT P. SARETT. *Nutrition Research Laboratory, Tulane University School of Medicine, New Orleans.* In eight normal persons on adequate diets of varied composition the urinary excretion of thiamine in 24 hours was over 100 μg., of riboflavin over 500 μg., and of N′-Methylnicotinamide (N′-Me) over 3.5 mg. In 4 hours after a test dose of thiamine (5 mg.), riboflavin (5 mg.) and niacinamide (50 mg.) mean thiamine excretion was 240 μg., riboflavin 2.3 mg., and N′-Me 6.4 mg. In seven normal persons on weighed diets providing 2500 calories, 40 gm. protein, 0.7 mg. thiamine, 0.5 mg. riboflavin and 6 mg. niacin, the urinary excretion of B vitamins decreased to relatively constant values, which averaged 19 μg. of thiamine, 56 μg. of riboflavin and 1.4 mg. of N′-Me. After three to six weeks on this diet the average four hour excretion in response to a test dose was 58 μg. of thiamine and 1.3 mg. of riboflavin. Excretion of N′-Me is not included in this report since the basal diet was supplemented with tryptophane before testing. In three patients with beriberi, thiamine excretion was 13 to 55 μg. in 24 hours and 21 to 55 μg. after a test dose. Of ten patients with B complex deficiency (glossitis, diarrhea, cheilosis, macrocytic anemia) two showed a thiamine excretion of less than 100 μg. in 24 hours and four an output of less than 60 μg. after a test dose. Riboflavin excretion in 24 hours was extremely variable (270 to 1800 μg.) and could not be correlated with test dose findings in appraising nutritional status. In 3 of the subjects excretion of N′-Me was less than 3 mg. in 24 hours and less than 3.5 mg. after a test dose. Nutritional evaluation based on excretion levels will be discussed.

The effect of threonine in choline deficiency. WENDELL H. GRIFFITH and MARY F. NAWROCKI (by invitation). *Department of Biological Chemistry, St. Louis University, School of Medicine.* The severity of renal hemorrhagic degeneration and the extent of deposition of liver fat are markedly increased in weanling rats by the addition of threonine to a low-choline, cystine-supplemented 8 per cent casein diet. No other essential amino acid shows a similar effect. Threonine is ineffective in the absence of supplementary cystine. The influence of threonine is not due to a direct antagonistic action on choline or on labile methyl but is the result of a stimulation of growth or metabolism which aggravates the existing choline deficiency. The effect of threonine is analogous to the previously demonstrated role of supplementary cystine in diets low in total sulfur amino acids (Mulford, D. J., and Griffith, W. H., J. Nutrition, **23:** 91 (1942)). This finding supports the conclusion that threonine and cystine are the most important limiting amino acids in casein fed at an 8 per cent level. If the 8 per cent casein diet is supplemented with cystine and threonine and with sufficient choline to permit survival, then other amino acids show apparent antilipotropic action. This experimental method provides a rapid and simple procedure for the determination of the priority of limiting amino acids in proteins. Application of the procedure to the determination of the relative metabolic activity of isomers and of derivatives of amino acids and to the recognition of sub-optimal intakes of vitamins and of other nutrients is in progress.

The effects of folic acid on respiratory metabolism. R. C. GRUBBS (by invitation), B. C. HOUGHTON (by invitation), JULIA TROSSBACH (by invitation) and F. A. HITCHCOCK. *The Laboratory of Aviation Physiology, The Ohio State University, Columbus.* Studies on respiratory metabolism and nitrogen balance before and during the administration of folic acid have been carried out on hospital patients amenable to folic acid and on normal controls. Oxygen consumption, carbon dioxide production and non-protein respiratory quotients were determined using the Tissot-Haldane technic under the following conditions: basal; one hour after a standard meal and before, during and after walking on a treadmill at a slow rate. All subjects lived in the hospital and were on a weighed diet. A two or three week control period was followed by a two or three week period during which 30 mgm. of folic acid were administered intramuscularly daily. Results indicate no significant change in basal oxygen consumption. There was an average increase of 11 per cent in the calories derived from carbohydrate in normals and of 6.5 per cent in patients. There was an increase in specific dynamic action in all patients and in approximately one-half of the normal subjects. In the nitrogen balance

studies, adequate diets were given and the caloric intake and nitrogen of the diet kept constant. An aliquot portion of the food was prepared each day for analysis. All urine and stools during the balance period were collected and total nitrogen determined by the Kheldahl method. Folic acid stimulated the appetite and increased nitrogen retention of patients but produced no consistent effect on the normals.

Factors affecting the excretion of metabolites of phenylalanine and tyrosine in alkaptanuria. W. KNOWLTON HALL (by invitation), V. P. SYDENSTRICKER and KATRINE RAWLS (by invitation). *Depts. of Medicine and of Biochemistry, Univ. of Georgia School of Medicine, Augusta.* One alkaptanuric adult female from a group of seven females and four males, all alkaptanuric and all related, was hospitalized for metabolic studies. The subject was fed a diet containing 70 gm. of protein per day and the urinary excretion of homogentisic acid, total nitrogen, organic acids, acetone bodies and creatinine were followed for a two week period and later for a three day period. The homogentisic acid excretion and homogentisic acid to nitrogen ratios were somewhat variable and were not significantly affected by the administration of an autolyzed yeast preparation, various types of oral and parenteral liver extracts or by six days on a ketogenic diet. As had been previously determined with other subjects with alkaptanuria from this group, phenolic compounds other than homogentisic acid were excreted in the urine. One or more of these compounds were precursors of melanin since on standing, 0.4 gm. or more of melanin per 24-hour specimen would form in the acidified urine, though there was no change in the homogentisic acid content. The total phenol rather than the homogentisic acid excretion roughly corresponded to the probable phenylalanine and tyrosine content of the diet. Similar results were obtained in a three day study of urine samples from a five year old alkaptanuric boy, apparently unrelated to the other alkaptanuric individuals. Studies were also made of factors influencing the excretion of homogentisic acid by rats fed a high tyrosine diet.

Dietary factors in experimental renal hypertension. I. Protein. PHILIP HANDLER and F. BERNHEIM (by invitation). *Departments of Biochemistry and of Physiology and Pharmacology, Duke University School of Medicine, Durham, N. C.* Series 1. Adult male rats, eating a commercial chow were rendered hypertensive by the Raska technic. Three weeks later, when systolic pressures were stabilized at 145–160 mm., they were divided into 3 groups of 12 rats each which were fed chow (A) and synthetic rations containing 50 (B) and 5% (C) casein. Pressures in group A remained at 155 mm. for 4 weeks and rose to 190–210 mm. before death (mean survival 51 days). Pressures in group B rose to 185–240 mm. within 1 week and all animals died within 4 weeks (mean 37 days). Pressures in group C fell to 95–130 mm. within 3 weeks, remained at that level for 2–5 weeks and then rose before death (mean survival 63 days). Series 2. Rats were fed synthetic rations containing 50 (D), 25 (E) and 5% (F) casein for 2 weeks before surgery and continued on these diets thereafter. Two weeks later mean systolic pressures were D 168 mm., E 141 mm., and F 114 mm. Pressures in group D rose to 180–220 mm. for 3 weeks and fell to subnormal levels in the last week as the animals lost weight. Pressures in group F remained low for 4 weeks and rose to 180–210 mm. the last week accompanied by a drastic weight loss. Group E remained at 140–155 mm. for 4 weeks and then behaved like group F. No correlation was observed between pressure and blood NPN or protein concentration. Studies are now in progress of the effects of 10% casein, choline deficiency, urea feeding, specific amino acids, and of salt feeding and deprivation on high and low protein diets.

Susceptibility to infection manifested by dogs on a low fat diet. ARILD E. HANSEN, OLETA BECK (by invitation) and HILDA F. WIESE (by invitation). *Dept. of Pediatrics, Univ. of Texas Medical School, Galveston.* Over a period of 4 years, it has been observed that young dogs maintained on a diet low in fat show an apparent lowered resistance to infection, especially of the skin. Of a total of 16 animals on the fat deficient diet, 9 have shown superficial impetigo-like infections of the skin. These have been resistant to external treatment. The skin of these animals also showed the desquamation characteristic for dogs on the low fat diet, whereas control animals receiving fresh lard in the diet quite uniformly manifest a healthy skin. The fat deficient animals become markedly emaciated and seem to be susceptible to pneumonia. Three of these dogs died and showed evidence of extensive pulmonary consolidation on autopsy. None of the control animals housed under identical conditions have shown any susceptibility to pneumonia. Especially significant has been the involvement of the ears in 10 animals. There has been a continuous purulent discharge from their ears. This condition has never developed in the control dogs. *Pseudomonas aeruginosa, alcaligenes fecalis,* paracolon and streptococci have been cultured from the purulent discharges. A striking cessation of the exudate from the infected ears occurs when fat is added to the diet. This has occurred when lard, bacon fat or butter fat have been substituted isocalorically for sucrose in the diet. Gain in weight, and a generally improved reaction of the animal together with the appearance of new healthy skin are also manifested to a greater or

lesser degree depending on the kind of fat which is added to the diet.

The effect of diet upon iron absorption. D. MARK HEGSTED, CLEMENT A. FINCH (by invitation), and THOMAS D. KINNEY (by invitation). *Department of Nutrition, Harvard School of Public Health; the Departments of Biological Chemistry, Medicine, and Pathology, Harvard Medical School; and the Medical Clinic and Department of Pathology, Peter Bent Brigham Hospital, Boston.* Modern studies on the absorption of iron from the intestinal tract have indicated that the amount absorbed is dependent upon the body needs for iron (Hahn, et al., J. Exp. Med. **78:** 169, 1943) and the work of Granick (J. Biol. Chem. **164:** 737, 1946) has supported the possibility that the control is by ferritin which acts as a block to iron absorption. However, the studies of Gillman, Mandelstam, and Gillman (S. Afr. J. Med. Sci. **10:** 109, 1945) and the work to be presented show that the absorption of iron is greatly influenced by the diet. Gillman et al. found a large proportion of South Africans to have excessive amounts of iron in their livers. We have shown that rats fed a diet composed chiefly of corn grits supplemented with iron deposit 10 to 20 times the normal amount of iron in the liver within a period of 4 to 6 weeks. Rats receiving adequate diets containing the same amount of iron show normal liver iron values. The excessive iron deposition is demonstrable histologically in amounts which parallel the chemically determined values and is found chiefly within the parenchymal liver cells. Stainable iron is also found in a few other organs when the liver values are high. The effect of various dietary supplements in preventing excessive iron absorption has been studied. None of the vitamins appeared to be active. Protein supplementation was only slightly effective while a complete salt mixture was more effective. Phosphate salts alone were similarly active but did not completely prevent a rise in liver iron.

Dental caries in the rat, mus norvegicus. JULIA O. HOLMES, L. R. Parkinson (by invitation), ANNE W. WERTZ (by invitation), and LOIS BROW (by invitation). *Agricultural Experiment Station, University of Massachusetts, Amherst, Massachusetts.* The study of tooth decay in the Albino rat has been continued, two strains of rats being used. The rations have been fed ad libitum for 14 weeks and have contained casein 15 to 24%, mineral mixture 4, fraction L-liver powder 4, corn oil 4, an adequate vitamin mixture, and either sucrose, glucose, dextrin or starch as the carbohydrate. Areas of yellow or opaque enamel or of yellow dentin, particularly soft areas, and decayed areas have been recorded as carious and the extent of decay has been scored on a scale of 1-2-3. On rations containing fermentable carbohydrates the average scores for incidence and extent of decay were 10 and 13, respectively, in strain I, and 19 and 36 in strain II. On diets containing starch or dextrin decay was less, the scores averaging 5 and 6 for strain I and 11 and 14 for strain II. Using basal rations of the above composition, several foods or biological compounds have been assayed for their caries-preventing potency. Urea prevented tooth decay. Some of the supplements increased the caries scores. The lowering of the casein level from 20 to 7% doubled the incidence and tripled the extent of decay. The data indicate that sugars are not the only components of the diet which may be conducive to tooth decay.

α-Tocopherol and certain nitrogenous compounds as factors influencing the mortality of rats after CCl_4 poisoning. E. L. HOVE. *Research Laboratories, Distillation Products, Inc., Rochester, New York.* Vitamin E and certain nitrogenous compounds have been found to influence the susceptibility of rats to carbon tetrachloride poisoning. The basal diet contained casein (10%), sucrose (76%), lard (10%), salt mixture, and an adequacy of pure vitamins, except vitamin E. Carbon tetrachloride was given intraperitoneally at a level of 2 cc. per kilogram body weight to groups of rats on this diet, with or without various supplements. The mortality of groups of rats on the basal diet was 90 to 100% within 48 hours after poisoning. Alpha-tocopherol (1 mg. daily) reduced the mortality to less than 20%. Supplements given for six weeks but stopped at the time of poisoning afforded several days protection but did not prevent ultimate death. Continuation of the supplements after poisoning gave continued protection. The activity of α-tocopherol was obliterated by 6-methyl thiouracil (0.1%), which had no activity by itself. Gamma-tocopherol was only slightly active at a level of 1 mg. daily. The protective action of α-tocopherol given to rats in the vitamin E free basal diet could be duplicated by methionine, xanthine derivatives, or other purines, or by an increased casein level in the diet. The influence of several individual amino acids has also been investigated. This technique is of value for the study of the intermediary metabolism of α-tocopherol and its several interrelations.

Relative importance of growth and metabolic rate on the utilization of vitamin A by the rat. R. M. JOHNSON (by invitation) and C. A. BAUMANN. *Department of Biochemistry, College of Agriculture, Univ. of Wisconsin, Madison.* Weanling rats were fed sufficient halibut liver oil to produce hepatic stores of approximately 250 gamma of vitamin A. They were then maintained on various low A diets designed to produce different rates of growth or different metabolic rates in the animals. The tissues of the animals were then analyzed at intervals. When growth was normal, the hepatic stores of vitamin A decreased and the concentration of vitamin in the kidney increased

The rate of decrease from the liver was accelerated slightly when the metabolic rate was high; it was decreased when the metabolic rate was low, e.g. when thiouracil was fed. Retention of liver stores was highest when growth was suppressed. In general the rate of utilization of stores vitamin A appeared to depend on both the growth rate and basal metabolic rate, but to a greater degree on the former.

The adequacy of an intake of seven milligrams of iron per day for women. FRANCES A. JOHNSTON, EUPHEMIA D. BOROUGHS, RUTH FRENCHMAN (introduced by HAZEL M. HAUCK). *N. Y. State College of Home Economics, Cornell University, Ithaca, N. Y.* Five young women were placed on a diet of 7 mg of iron for a four-week period. Seven milligrams was selected because Leverton (J. Nutrition **21:** 617, 1941) had found retentions were large enough to cover all losses on that intake. In this study, when the amounts of iron lost in urine and stools were considered, retentions were 0.64, 0.26, 0.43, 0.61, 1.33 mg per day. The menstrual losses per period ranged from 10.27 to 28.08 mg. In one case the retention was more than enough to cover menstrual losses and in four cases the retentions almost equalled the losses. Those subjects whose menstrual losses approximated their retentions were probably not obtaining sufficient iron to cover all their needs because no account was taken of iron lost in perspiration, skin, nails and hair. The conclusion drawn from this study was that an intake of seven milligrams of iron was on the borderline of the requirement for four of these five women when the diet was adequate in other respects and contained one small serving of a low iron flesh food. It was below the requirement if the iron content of perspiration is an appreciable amount. When beef containing 3.4 mg of iron was added to the diet, retentions ranged from 1.77 to 2.53 mg per day. This high retention with beef may explain the better retentions found by Leverton than by us on the 7 mg intake, as the diet she used contained much beef.

The inhibitory effect of excess methionine on protein utilization. CHARLES F. KADE, JR. and JESSY SHEPERD (introduced by AARON ARNOLD). *From Biology Division, Sterling-Winthrop Research Institute, Rensselaer, N. Y.* It is well known that the growth rate of the rat can be increased by the addition of small amounts of methionine to a diet containing suboptimal quantities of casein. Studies were made to determine the effect of supplements of methionine on the growth of rats fed diets containing casein at an intake giving the highest protein efficiency. It was found that a diet containing 8 per cent casein (based on nitrogen × 6.25) permits the greatest utilization of the protein. Varying quantities of DL-methionine up to 1 per cent added to this diet resulted in increasing protein utilization. An 8 per cent casein diet supplemented with 1.5 per cent DL-methionine gave little better growth than the diet without additional methionine; 2 per cent, 2.5 per cent and 3 per cent added methionine definitely inhibited growth and protein utilization. In order to rule out taste factors and differences in caloric intakes a set of weanling rats were pair fed isocaloric diets differing only in the DL-methionine content. The rats fed the 8 per cent casein diet supplemented with 1.0 per cent DL-methionine grew the best, showing the highest protein efficiency. Two per cent DL-methionine added to casein was little better than the unsupplemented casein. Thus greater than optimum quantities of methionine are of no value in improving the utilization of casein and may actually be inhibitory. In all probability the mixture of amino acids in which the proportion most nearly approaches the quantitative requirements of the animal for the essential amino acids will be the most efficiently utilized.

Effect of xanthophyll on the utilization of carotene and vitamin A by the rat. BARBARA KELLEY (by invitation) and HARRY G. DAY. *Dept. of Chemistry, Indiana Univ., Bloomington.* Weanling rats were made vitamin A deficient by standard procedures. After growth had nearly ceased they were divided into similar groups as respects weight and sex. In each experiment β-carotene in cottonseed oil was given daily to one group, the amounts ranging from 4 to 20 γ per supplement in the different experiments. A second group was given exactly the same amount of carotene plus 20 to 100 times as much xanthophyll (lutein) in equal amounts of cottonseed oil. A third group received xanthophyll alone and the fourth received neither carotene nor xanthophyll. The supplements were given for 21 to 28 days. In all the experiments rats receiving carotene alone gained more weight than those given xanthophyll in addition to the carotene, although the gains were quite variable. The amount of vitamin A in the livers was decidedly greater in the rats given carotene alone. Also, marked differences were found when vitamin A (D.P.I. concentrate) was given as the supplement and relatively large amounts of xanthophyll were added. This was particularly notable with regard to the effect on vitamin A in the kidneys. Rats receiving xanthophyll plus vitamin A had less than one-half as much of the vitamin in the kidneys. The results suggest that the effect of non-provitamin A xanthophyll on growth and the vitamin A content of the tissues is not due to a specific impairment of the enzymatic mechanism for the conversion of carotene into vitamin A.

Biological analysis of rice. MARINUS C. KIK. *Department of Agricultural Chemistry, University of Arkansas, Fayetteville.* Studies were continued to reveal the losses of vitamins in the regular

practice of commercial rice milling. Two series of rat feeding experiments were initiated to learn whether whole brown rice was a better source of biotin and folic acid than white polished rice. *Biotin.* In triad feeding, using 12 triads, no differences in gain were found and no increased growth was obtained from whole-brown-rice-rations to which biotin was added. A slightly better growth was obtained on the whole-brown-rice-rations in paired feeding tests. *Folic Acid.* In 12 triads, polished rice was inferior to whole brown rice as a source of folic acid. The brown rice ration was not improved by the addition of more folic acid. No difference was found in paired feeding tests. In these experiments, all rats received daily 20 μg of thiamine, 20 μg of riboflavin, a 20 μg of pyridoxine, 6 mg of choline, 120 μg of Ca pantothenate, 10 mg of inositol, 3 mg of para-amino-benzoic acid, 2.5 μg of biotin and 5 μg of folic acid. In addition, each animal received 3 drops per week of halibut liver oil as supplementary sources of vitamins A and D. The vitamin to be tested was omitted.

In rats fed white polished rice diets with and without *niacin*, the same growth was obtained. A similar experience was obtained with rats fed brown rice diets. In each case 12 pairs of rats were used in paired feeding experiments. This suggests that white polished rice and whole brown rice, unlike corn or corn grits, do not have any growth retarding effect in rats.

The nutritive value of cereal proteins in human subjects. CARL A. KUETHER (by invitation) and VICTOR C. MYERS. *Department of Biochemistry, Western Reserve University, Cleveland.* Nitrogen balances were measured in eight adult human subjects (3 F, 5 M) eating weighed diets composed of cereal, milk, cream, sucrose, butter and vitamin capsules. The diets were designed to provide 0.7 gm of protein (N × 6.25) and approximately 37.5 Cal per kilo per day, with the cereal providing 55, 80 or 100 per cent of the protein. The cereals used were both processed and unprocessed oats, wheat and corn. The nitrogen of the exploded oat cereal (Cheerioats) was utilized as well as the nitrogen of rolled oats (mean replacement value, 101 per cent) under these conditions. Supplementation with DL-lysine improved the nitrogen balance in every case, the extent of the improvement being the same for both of these cereals. Both wheat flakes and exploded wheat were inferior to whole wheat in maintaining nitrogen balance (mean replacement values of 95 and 84 per cent, resp.), indicating damage to the protein in the processing of these cereals. Corn flakes and an expanded corn cereal (Kix) were approximately equal to whole corn in maintaining nitrogen equilibrium (mean replacement values of 99 and 101 per cent, resp.) under the conditions of this experiment. Three of the male subjects were on all of the diets where the cereal supplied 100 per cent of the protein. The mean daily nitrogen balances for these three subjects, arranged in the order of decreasing nitrogen balance, were as follows: whole wheat, −0.71 gm.; exploded oat cereal, −1.17; wheat flakes, −1.19; rolled oats, −1.22; expanded corn cereal, −1.55; whole corn, −1.60; corn flakes, −1.67; exploded wheat, −2.22.

The quantitative requirement of the rat for magnesium and effects of magnesium deficiency in the rabbit. H. O. KUNKEL (by invitation) and P. B. PEARSON. *Dept. of Biochemistry and Nutrition, A & M College of Texas, College Station.* Diets containing different levels of magnesium ranging from 50 to 300 p.p.m. were fed to growing rats. When the magnesium is provided as the sulfate, between 150 p.p.m. and 200 p.p.m. is required to maintain normal blood magnesium values. Slightly lower values may be sufficient to protect the animal against vasodilatation and hyperexcitability. The magnesium in dried wheat plant or as the oxide or carbonate is less efficiently utilized than magnesium sulfate. Weanling rabbits fed a diet deficient in magnesium exhibit within a period of 5 to 6 weeks a syndrome of hyperexcitability, convulsions, hypomagnesemia and retardation of growth. The addition of magnesium to the diet of rabbits that have ceased to grow results in a prompt resumption of growth. The vasodilatation characteristic of a magnesium deficiency in rats was not observed in rabbits. The magnesium content of the whole blood of rabbits on an adequate diet is approximately 4.8 mg. % while the level in the plasma is about 1.6 mg. %. The plasma magnesium reaches a minimum level of about 1 mg. % after six weeks on the deficient diet. The magnesium content of whole blood continues to decline over a longer period of time to a level of approximately 3.4 mg. % after ten weeks.

Urinary thiamine and riboflavin excretion of children during fasting and under conditions of loading test. WANDA I. LAMECK (by invitation), MARGARET N. CORYELL (by invitation), ELIOT F. BEACH and ICIE G. MACY. *Research Laboratory, Children's Fund of Michigan.* During a survey of nutritional status in five institutions the urinary excertion of thiamine and riboflavin by 374 children was determined during fasting and for 4 hours following an oral dose of 1 mg. of each of these vitamins. Fluorophotometric methods were used for analysis. The accuracy of short period urine collections was verified by simultaneous creatinine and total nitrogen determinations. The data reveal that in children three to seventeen years old, age and size have little effect either on hourly fasting excretion of B vitamins or response to load tests. In most cases boys showed a higher average excretion than girls, perhaps owing to the greater food consumption. Comparison of the fasting and load

test excretion data for 5 institutions reveals a considerable spread in values which is believed to indicate differences in level of tissue saturation with respect to thiamine and riboflavin. Average fasting hourly thiamine excretions varied from 4.3 to 9.7 μg. and 4 hour load test outputs ranged from 104 to 189 μg. Corresponding values for riboflavin were 32 to 55, and 273 to 387 μg, respectively. With either vitamin, correlation between fasting and load test values was low. Between spring and fall, average excretion of thiamine rose from 5.7 to 8.3 μg. per hour, apparently due in part to seasonal changes in dietary habit. In one group of children a period of dietary improvement was followed by an increase in the average fasting excretion of riboflavin from 39 to 48 μg. per hour.

Diet and dose-response of weanling rats to intravenous alloxan. GEORGE V. MANN (by invitation) and FREDRICK J. STARE. *Department of Nutrition, Harvard School of Public Health, and Department of Biological Chemistry, Harvard Medical School, Boston.* The relationship of the diabetic response in weanling rats of several strains has been determined by measurement of the degree and duration of diabetes in animals after a single intravenous dose of alloxan monohydrate. Blood sugar levels were measured after a 5-hour fast. Urine sugar was determined on 24-hour urine collections. A fasting blood sugar above 150 mg. per cent and 24-hour urine glucose above 0.1 gm. was considered as an indication of diabetes. A technique has been developed with which animals can be injected rapidly and accurately through a tail vein without anesthesia. The following table indicates the diabetic response with varying dosage levels of alloxan:

The diabetic response of young rats after intravenous alloxan

Dosage	No. of rats	48-hour Response	Immediate mortality
mg./kg. body wt.		%	%
30	10	10	0
40	10	40	0
50	9	70	0
60	10	90	0
70	32	97	7*

* Due to toxic effects of alloxan upon liver and kidneys.

The fluctuation of incidence of diabetes after a given dose of alloxan has been found to be related to the nature of the diet fed. It has been found that isocaloric substitution of carbohydrate with protein increases the survival rate of the animals. Substitution of 1% sodium chloride for drinking water lowers the survival rate irrespective of the protein level of the diet. A natural diet (Gaines Dog Meal) with 25% protein allows a higher survival rate than a synthetic diet containing 45% protein.

Food fermentation as a preventive of furunculosis. J. F. MCCLENDON. *Hahnemann Medical College, Phila.* With the development of canning and soft drinks, the eating and drinking of food products preserved by fermentation has decreased whereas the percentage of calories from sugar and bread have increased, especially during the meat shortage. There are mild diabetics whose only symptom is furunculosis. One such was observed for the last 30 years in relation to the effect of diet, insulin, sulfanilamide, sulfathiazole and penicillin. A high fat-meat diet was tried for a few years, then a low available-sugar diet for 2 years and a low cellulose-hemicellulose diet for a number of years. Since it was found that acetate injected into the blood stream is rapidly metabolized and since acetate is produced and absorbed by the 50-litre rumen of a sheep, a 50-litre crock was used in the fermentation of human food. Since the furunculosis was seasonal, i.e. during the summer when outdoor work and fresh fruits and vegetables stimulated the appetite, the experiments were confined chiefly to the removal of sugar from the food by fermentation and the substitution of pickles for part of the bread of the diet. (On a diet containing coarse corn and leafy vegetables considerable fermentation occurs in the gut and it seems probable that thiamin, riboflavin, pantothenic, nicotinic and folic acids and biotin may be synthesized by bacteria in the gut and absorbed from dead bacteria.) This diet shrinks the skin and reduces body weight and prevents furunculosis. Return to a high available carbohydrate diet reverses all these effects.

Calcium requirements of seven adolescent girls. BEULA V. MCKAY (by invitation), MRS. ELEANOR SMITH (by invitation), and JANICE M. SMITH. *Agricultural Experiment Station, Dept. of Home Economics, University of Illinois, Urbana, Illinois.* Seven girls, 12–13 years of age, selected on the basis of physiological maturity and willingness to cooperate served as subjects. Continuous calcium, nitrogen and phosphorus balance determinations were made for 79 days during the summer of 1946. Four levels of calcium were fed during consecutive 20-day periods. The basal diet, composed of a variety of natural foods, furnished, on the average, 183 mg. calcium daily. To this were added homogenized milk and ice cream in quantities to supply the following average amounts of calcium daily: Period I, 1.995; Period II, 2.303; Period III, 0.935 and Period IV, 0.455 grams. The average daily retention for the seven girls was as follows: Period I, 0.420; Period II, 0.463; Period III, 0.224 and Period IV, 0.035 grams. Individual data were used to determine values for utilization of milk calcium and resulting daily calcium requirements according to the technique of Holmes et al (J. Nutrition **31**: 127, 1946). The percentage utilization of calcium from milk and ice cream was 23.1, 39.0, 40.1, 30.3,

34.1, 40.0 and 54.2 for each of the seven subjects. Four of the girls showed maximum calcium retentions and their calculated daily calcium requirements were 1.009, 1.654, 1.113 and 0.700 grams. The low figure resulted from the observed high utilization value of 54.2%, not from a lowered retention. One of the the three remaining girls did not exhibit maximum calcium retention and the other two were doubtful. Nitrogen and phosphorus data will be reported later.

Effect of arginine on urinary output of 17-ketosteroids in a patient with myotonia atrophica. A. T. MILHORAT. *Depts. of Psychiatry and Medicine, Cornell Univ. Medical College, the Russell Sage Inst. of Pathology and the New York Hospital, New York, N. Y.* A man aged 46 with moderately advanced myotonia atrophica showed generalized muscular wasting and advanced testicular atrophy. The average daily urinary outputs of preformed creatinine and creatine were 0.750 and 0.2000 gm. respectively. The BMR was −26 per cent. The concentration of free cholesterol in the blood serum was 183 mg. per cut; the level of cholesterol esters was 467. Examination of the semen showed azoospermia. The daily urinary output of 17 Ketosteroids was only 3.66 mg. Delta-tocopherol in dosages of 500 mg. daily for a period of 7 weeks induced no demonstrable changes except an increase in androgen-output to a level of 5.95 mg. Administration of delta-tocopherol was stopped and L (+) arginine mono-hydrochloride was given daily in amounts of 2 gm. for 4 days, 6 gm. for 6 days, and subsequently in amounts of 12 gm. daily. On the 9th day of arginine feeding the urinary output of 17 Ketosteroids was 6.30 mg. and on the 21st day the output had increased to 8.72 mg. No effect on output of preformed creatinine and creatine, BMR, or blood cholesterol was observed. When considered together with the observations of Kochakian and Fraenkel-Conrat, Simpson and Evans that testicular and adrenal cortical hormones increase arginase activity, these findings suggest that arginine and arginase are important components of the mechanism regulating production of these hormones.

Studies of the occurence of diketo-l-gulonic acid, dehydro-l-ascorbic acid and l-ascorbic acid. MARY B. MILLS (by invitation), CHARLOTTE M. DAMRON (by invitation), and JOSEPH H. ROE. *Department of Biochemistry, School of Medicine, George Washington University, Washington, D. C.* The 2:4-dinitrophenylhydrazine procedure has been made the basis of a method for the determination of diketogulonic acid, dehydroascorbic acid and ascorbic acid in the presence of each other. A difficulty is the extraction of tissues without producing a change in the partition of the three compounds. This is overcome by grinding the tissues under 10 per cent $SnCl_2$ in 5 per cent HPO_3 solution, then diluting the slurry with 5 per cent HPO_3 solution until the $SnCl_2$ has a final concentration of 0.5 per cent. This method has been applied to the analysis of plant and animal tissues. Diketo gulonic acid does not occur in the tissues of the normal guinea pig in amounts determinable by this method. Dehydroascorbic acid has been observed in very small amounts in guinea pig tissues but it is uncertain whether this is a true finding or the result of oxidation of ascorbic acid during extraction. Fresh foods from the market may show the presence of small amounts of dehydroascorbic acid or they may not contain any of this compound. Processed and dehydrated foods usually show the presence of small quantities of diketogulonic acid and considerable amounts of dehydroascorbic acid. The gradual loss of total ascorbic acid and the rate of change in the partition of ascorbic acid, dehydroascorbic acid and diketogulonic acid in kale, orange juice, and potato slurry in the ice box have been studied.

Inadequacy of thymine as a nutritional factor for reproduction and lactation in the mouse. LEONORA MIRONE (by invitation) and LEOPOLD R. CERECEDO. *Department of Biochemistry, Fordham University.* The ability of thymine to substitute for pteroylglutamic acid in reproduction and lactation in the mouse was investigated in this laboratory. This seemed of interest since Spies, T. D. et al. (Southern Med. J., **39:** 269, 1946) reported that thymine was beneficial in human anemias. To the basal diet R-5(a) 6 grams of thymine (diet RT-5) were added in lieu of 10 mg. of pteroylglutamic acid (diet RS-5(10)). Sixteen females which had been maintained on the basal diet R-5(a) since weaning and which represented three different strains of mice were fed diet RT-5 at mating. The results obtained are summarized below.

Diet	Successful gestation	Litters weaned	Litter size	Weaning weight
	%	%		g.
R-5(a)	61	33	5.5	7.5
RT-5	70	45	6.3	6.7
RS-5(10)	82	70	6.3	8.5

The findings indicate that thymine can not substitute for pteroylglutamic acid as a nutritional factor for reproduction and lactation in the mouse when fed during gestation and lactation. Recently Petering, H. G. and Delor (Science, **105:** 547, 1947) reported the inability of thymine to substitute for pteroylglutamic acid in the rat.

Studies on fat digestibility; the effect of melting points and dietary calcium and magnesium levels. MARGARET G. MOREHOUSE (by invitation), AMBER LIENG-SHAN CHENG (by invitation) and HARRY J. DEUEL, JR. *Univ. of Southern California School*

of Medicine. Previous studies on fat digestibility (Crockett, M. E., and H. J. Deuel, Jr., J. Nutr., 33: 187, 1947) have shown that with melting point increase above 50°C. a decrease in total digestibility occurs with an increase in the per cent of soap in the unabsorbed fat residue. The present investigation is a study of the effect of calcium and magnesium in the diet on the digestibility of natural and synthetic fats of various melting points. The food consumption of female rats maintained on diets containing 15% fat with or without added calcium and magnesium was recorded. The fat digested was measured by subtracting neutral fat, fatty acids and soaps extracted from fecal residues from the fat consumed. Corrections for endogenous fecal fat were made. The digestibility of partially hydrogenated lard (M. P. 55) decreased from 75 without to 60% with the inclusion of calcium and magnesium. In the latter case a large proportion of the unabsorbed fat appeared as soaps. The digestibility of a blended lard (M. P. 48) averaged 95% and was independent of the dietary salt levels. However, the digestibility of synthetic triglycerides melting at this level decreased with the addition of the divalent ions. Trilaurin (M. P. 47) averaged 95% digestibility without and only 71% with the salt addition. The corresponding changes in the digestibility of trimyristin, tripalmitin, and tristearin were 77 to 38, 28 to 14, and 19 to 11 per cents without and with the salts respectively.

Studies of thiamine deficiency in C3H mice. HAROLD P. MORRIS, CELIA S. DUBNIK, and THELMA B. DUNN. *National Cancer Institute, U.S.P.H.S., Bethesda, Md.* The addition of a small amount of thiamine to a diet devoid of thiamine resulted in relative good growth of young mice for a few weeks, followed by a decline in weight, development of clinical symptoms of thiamine deficiency which terminated in death of the animal unless the mice received thiamine supplements. This syndrome has been designated chronic thiamine deficiency. Gross and histological changes were observed in mice subject to this slowly produced thiamine deficiency. The changes were characterized by an absence of spermatogenesis, by muscular degeneration and by hemorrhages in the mid-brain but by an absence of histologic changes in other parts of the nervous system. The relation of enforced food restriction on survival, body weight and other changes observed in chronic thiamine deficiency will be discussed. Some quantitative values for the amount of thiamine required for growth and maintenance of body weight will be given for C3H mice.

Composition of Central American foods. I. Honduras. HAZEL E. MUNSELL, ROBERT S. HARRIS and LOUIS O. WILLIAMS (by invitation), with the technical assistance of LOUISE GUILD (by invitation), GERTRUDE NIGHTINGALE (by invitation) and CYNTHIA TROESCHER (by invitation). *Nutritional Biochemistry Laboratories, Massachusetts Institute of Technology.* For the establishment of a sound program of food production and food consumption it is important to know the composition of the indigenous foods which are, or can be, grown in the area. On this basis a long range study of the composition of foods grown in Central America is underway. Specimens are collected and identified by a botanist, samples representative of the edible portion are stabilized and shipped by air express to Cambridge. Whenever possible a record is made of the soil type, annual rainfall, altitude and whether the sample was grown on fertilized land. A comparison of the data from similar samples may give information on the relative importance of genetic and non-genetic factors in determining the composition of edible plants. Moisture, nitrogen, ether soluble fraction, crude fiber, ash, calcium, phosphorus, iron, carotene, thiamine, riboflavin, niacin and total ascorbic acid content are being measured. In the present report the results of analyses of 129 samples of 85 plants collected in Honduras are presented.

Response of leukopenia and granulocytopenia in sulfathiazole-fed rats to pteroylglutamic acid and to parenteral liver extracts. CHARLES W. MUSHETT (by invitation) and GLADYS A. EMERSON. *Merck Institute for Therapeutic Research, Rahway, N. J.* Treatment of rats, which had developed leukopenia and granulocytopenia while maintained on a purified diet containing 1% sulfathiazole, with either pteroylglutamic acid or purified liver extracts containing the antipernicious anemia factor resulted in a prompt restoration to normal of the white cell numbers. A significant increase in reticulocytes was observed also. Differential counts on femoral bone marrow of rats treated with the liver preparations revealed a picture similar to that seen in animals given pteroylglutamic acid. In contrast to the untreated rats which had a predominance of erythroid elements in the bone marrow and few or no polymorphonuclear leukocytes, the treated animals had relatively more myeloid cells, including a large number of segmented neutrophiles. This resulted in an increased myeloid-erythroid ratio. Microbiological assays of the liver extracts failed to show the presence of a sufficient amount of pteroylglutamic acid or its conjugates to account for the hematopoietic response observed. The leukopenia and granulocytopenia found in rats given a purified diet of low protein content, similar to that used by Daft could also be corrected by injections of the highly purified liver extracts.

Nutrients required by the rat for reproduction and lactation. B. L. O'DELL (by invitation) and A. G. HOGAN. *Department of Agricultural Chem-*

istry, University of Missouri, Columbia. Rats from the stock colony were placed on a synthetic ration composed of acid washed casein, cerelose, wood pulp, salts, lard, vitamins A, D, E, and K, thiamine, riboflavin, pyridoxine, and calcium pantothenate. All of the surviving offspring were allowed to remain with the dam for four weeks. On the basal ration 48 per cent of the young were weaned with an average weight of 42.2 g. The weaning percentages and weaning weights resulting from the addition of various vitamin combinations were as follows: Niacin, 34%, 37.6 g.; choline and niacin, 51%, 48 g.; biotin, choline, and niacin, 66%, 46.9 g.; pteroylglutamic acid, biotin, choline, niacin and ascorbic acid, 75%, 56.4 g.; inositol, pteroylglutamic acid, biotin, choline, niacin and ascorbic acid, 71%, 56.3 g.; inositol, biotin, choline, niacin, and ascorbic acid, 62%, 53.2 g. When the casein was extracted with alcohol the response was 74%, 52.6 g.; when lactalbumin was substituted for casein the response was 66%, 51.0 g. In other trials the amount of vitamin A in the diets varied from 100 to 2000 I. U. per 100 g. and the amount of vitamin E from 0.5 to 2.5 mg. Of 1628 young rats born to dams receiving pteroylglutamic acid, only 2 or 0.12% developed hydrocephalus. Of 2134 young born to dams which did not receive the vitamin, 22 or 1.03% developed hydrocephalus. There was no correlation between any of the other dietary modifications mentioned and the incidence of hydrocephalus in the young.

Haemoglobin levels by age and sex. L. B. Pett and G. F. Ogilvie (by invitation). *Nutrition Division, Department of National Health and Welfare, Ottawa, Canada.* Although haemoglobin estimations have been used for a century and are important in nutrition surveys, data are difficult to interpret for several reasons: (1) methods in common use differ widely in their accuracy, some being quite unsuitable; (2) results expressed in percentages may not be clear since at least 5 different percentage standards are in use; (3) there is no really "normal" value and little information is available on ranges to be expected in a mixed population, especially at different ages. In the course of doing nutrition surveys in many parts of Canada, thousands of haemoglobin estimations have been made on people of all ages, using one photoelectric apparatus. This paper reports some of the sources of error involved in haemoglobin estimations generally, and the results tabulated by age and sex of 3,148 estimations from age 1 to 79. While the averages found are not suggested as necessarily "normal", yet they represent the most widespread collection of figures in Canada on unselected people, obtained by the same procedure. As has been shown before, male haemoglobin levels gradually rise to a peak about age 20; in this report the peak average was 14.5 grams. Female levels showed no such consistent rise with age, although the averages found for different age groups varied from 12.3 grams to 13.2. The use is advocated of distribution curves rather than averages or percentages in recording low haemoglobins for surveys.

The effect of vitamin therapy upon serum blood levels. H. B. Pierce, R. F. Krause (by invitation), J. H. Browe (by invitation), Susan Merrow (by invitation) with the technical assistance of C. A. Newhall (by invitation), T. H. Harwood (by invitation), Hattie Kaplan (by invitation), Anne Baker (by invitation), Jean Parker (by invitation), Lorraine Carpenter (by invitation). *Departments of Biochemistry, Medicine and Anatomy, College of Medicine, University of Vermont, Burlington, Vermont.* Vitamin therapy has been used for one year with a group of school children having all degrees of several lesions assigned to a deficiency of one or more vitamins. Niacinamide, riboflavin, ascorbic acid and vitamin A are being given to treat lesions generally attributed to their respective deficiencies. Blood samples have been analyzed three times during the year, in winter, spring and fall, according to the microchemical methods of Bessey and associates. Therapy with vitamin C resulted in a rapid and marked increase in serum ascorbic acid levels; however, in vitamin A therapy, serum levels of this vitamin did not rise as rapidly or as high proportionately. The percentage of children on A therapy showing increased blood levels of vitamin A was less than that noted for vitamin C in the ascorbic acid group. In all therapy groups serum ascorbic acid and vitamin A levels are higher than in control groups regardless of the vitamin used for therapy. Control groups continue to show a high percentage of children with low serum values of ascorbic acid, vitamin A and carotene. Computations derived from food intakes show that greatest inadequacies in the diet are vitamin A and ascorbic acid.

Pantothenic acid requirement of adrenalectomized rats and its relation to the white blood cells. Elaine P. Ralli, Mary E. Dumm (by invitation), and Paul Roth (by invitation). *Department of Medicine, New York University College of Medicine.* Previous observations (Ralli, *Endocrinology*, 1946) have shown that following adrenalectomy in rats survival was greatly prolonged when large amounts of pantothenic acid were added to the diet (4 to 6 mg. daily). The present studies are concerned with establishing the minimal dose of pantothenic acid required for prolonged survival. Observations have also been made on the total white blood cell count and the per cent of lymphocyte and polymorphonuclear cells in normal rats on the diet deficient in pantothenic acid and in rats after adrenalectomy when the diet was either deficient or supplemented with varying amounts

of pantothenic acid. At the present time the doses of pantothenic acid that have been tested are .03 mg., 1 mg., and 2 mg. daily. On the first two amounts the mean survival was 14 days, with a S.D. ± 4.3, and 13 days with a S.D. ± 1.9, respectively. On 2 mg. 50 per cent of the rats have survived more than 20 days. This dose is apparently not as effective as the original massive doses used (4 to 6 mg. daily). Further experiments on graded doses of pantothenic acid are in progress. The total white blood cell count increased after adrenalectomy to a significant extent in the rats receiving pantothenic acid. There was also an increase in the white blood cells in the adrenalectomized deficient animals, but this was not statistically significant. The total lymphocytes per cubic millimeter were significantly increased in all groups following adrenalectomy.

Southern peas as a source of protein for growth. L. R. RICHARDSON. *Department of Biochemistry and Nutrition, A & M College of Texas, College Station.* Southern peas (table varieties of the cowpea) were compared with English peas, pinto beans, lima beans, egg albumen and casein as a source of protein for growth for rats. The legumes furnished all the protein in a purified diet at a level of 10% protein. The vitamins were supplied as an A-D concentrate and as pure compounds. There were 8 rats in each test and the experimental period was four weeks. Cooking did not improve either the Southern peas or the English peas as the same gains were obtained with the raw peas as with the cooked peas in each case. However, the rats fed English peas gained twice as much as those fed Southern peas. Those fed raw lima beans and raw pinto beans lost 12 to 16 grams, while those fed cooked lima beans and cooked pinto beans gained 25 and 10 grams, respectively. The gain on Southern peas was equal to that of the rats fed cooked lima beans. Supplementing the cooked legumes with 0.2% of *DL*-methionine increased the gains 2 to 3 times. The average gains were 76, 72, 52 and 46 grams, respectively, when lima beans, English peas, pinto beans and Southern peas were supplemented with methionine. In tests with Southern peas and pinto beans 0.1% of methionine was inadequate and 0.3% was no better than 0.2%. The gain was 56 grams when the diet contained 10% of egg albumen, 34 grams when it contained 10% casein and 96 grams when it contained 20% casein.

The riboflavin content of the diet and leg trouble in broad breasted bronze turkeys. L. R. RICHARDSON, R. M. SHERWOOD (by invitation), H. L. GERMAN (by invitation), *Departments of Biochemistry and Nutrition and Poultry Husbandry, A & M College of Texas, College Station, Texas.* The effect of a border line amount of riboflavin on leg disorders in broad breasted Bronze turkeys was tested with four diets. The constant components of Diets A, B and C were yellow corn meal, wheat shorts, wheat bran, soybean meal, bone meal, vitamins A and D, manganese sulfate and choline. Crystalline riboflavin was added so that Diet A contained an average total of 1.68 mg. of riboflavin per pound. Diet B contained 1.33 mg. and Diet C contained 2.5 mg. Diet D was high in animal protein and contained an average of 2.04 mg. of riboflavin per pound. A total of 7 out of 31 or 22.6 per cent of the turkeys fed Diet A, and 9 out of 38 or 23.7 per cent fed Diet B developed some form of leg trouble by the end of the 23rd week. The symptoms appeared first when the turkeys were 13 weeks old. There were no leg abnormalities in the 64 turkeys which received Diet C or D. Seven of the affected birds were each given 9 doses of 5.0 mg. of riboflavin. One did not recover. One recovered in 2 days and the other five recovered in 3 to 4 weeks. Commercial feeds from 2 farms where there was a severe outbreak of leg trouble contained 1.49 and 1.78 mg. of riboflavin per pound. These data together with those obtained in the laboratory indicate that leg trouble in turkeys may be due in part to an inadequate supply of riboflavin.

The effect of pantothenic acid deficiency on acetylation in rats. THOMAS R. RIGGS (by invitation) and D. MARK HEGSTED. *Dept. of Nutrition, Harvard School of Public Health and Dept. of Biological Chemistry, Harvard Medical School, Boston.* Lipmann *et al.* have studied *in vitro* acetylations of choline and sulfonilamide, and have concentrated the coenzyme (designated coenzyme A) necessary for the reaction (J. Biol. Chem., **160:** 173 (1945); **162:** 743 (1946); Federation Proc., **5:** 145 (1946)). Since these concentrates were found to contain large amounts of pantothenic acid (J. Biol. Chem., **167:** 869 (1947)) the present studies were undertaken to demonstrate the need of this vitamin by rats for normal acetylations *in vivo*. p-Aminobenzoic acid (PAB) was used as the test substance throughout. Normal rats were found to acetylate nearly 70% of the amount excreted in 24 hours of a 1 mg. or 2½ mg. dose of PAB administered intraperitoneally. Animals rendered pantothenic acid deficient acetylated only 50% of a 1 mg. dose and less than 40% of a 2½ mg. dose. Simultaneous injection of 1 mg. of calcium pantothenate to deficient animals immediately returned their acetylation to normal. Investigations also were made on the effect on acetylation of added acetate, 24-hour fasting, size of PAB dose, and of thiamine and riboflavin deficiencies.

Blood constituents of swine in a pantothenic acid deficient condition. WALTER C. RUSSELL and ARTHUR E. TEERI (now at the University of New Hampshire, Durham, New Hampshire). *Department of Agricultural Biochemistry, New Jersey Agricultural Experiment Station, Rutgers University, New Brunswick, New Jersey.* In an attempt

to characterize the pantothenic acid deficiency in swine, determinations were made of certain nitrogenous, lipid, and inorganic constituents of the blood. In comparison with the positive controls, creatinine and serum sodium of the deficient animals showed a tendency toward lower values and for serum chloride, a lower trend was evident in some animals but not in others. Creatine and serum protein values were higher for the deficient animals. For both groups, blood potassium showed a downward trend with age, the value becoming lower for the deficient animals as the deficiency developed. Following an initial rise for both the control and deficient groups, the plasma levels of total lipids, total cholesterol, cholesterol esters, and free cholesterol of the deficient animals became lower at 80 to 90 days of age and remained so throughout the experiment. Between 105 and 164 days of age, the close of the experiment, the average values for total lipids, total cholesterol, cholesterol esters and free cholesterol were 35, 37, 34, and 57% less, respectively, than those of control animals. The significance of these observations in the metabolism of the domestic pig will be discussed.

The tryptophane requirement for growth of the rat. W. D. SALMON. *Laby. of Animal Nutrition, Alabama Polytechnic Inst., Auburn.* The minimum tryptophane required for a normal rate of growth of the Alabama Agricultural Experiment Station (AES) strain of rats was determined. The basal diet contained tryptophane-free casein hydrolysate equivalent to 12% of casein, corn grits 40%, salts 4%, cystine 0.3%, choline chloride 0.2%, sucrose to 100%, and the known essential vitamins except nicotinic acid. The addition of 0.2% DL-tryptophane made the diet adequate for normal growth; 0.18% was inadequate. When 20 mg. nicotinic acid was added per kg. of diet, 0.1% DL-tryptophane was adequate for normal growth. When 9% of casein was substituted for the casein hydrolysate in the basal diet, 0.06% DL-tryptophane or a total tryptophane content of 0.205% was adequate for normal growth in the absence of nicotinic acid. However, 15% of casein in the diet did not furnish sufficient tryptophane for normal growth in the absence of nicotinic acid, although this amount of casein supplied 0.235% tryptophane. Moreover, when nicotinic acid was added to the casein hydrolysate diet, supplementation with 0.1% DL-tryptophane was more effective than the same amount of tryptophane supplied in the form of casein.

Effects of gelatin, glycine and pyridoxine on tryptophane and nicotinic acid metabolism in humans. HERBERT P. SARETT and GRACE A. GOLDSMITH. *Nutrition Research Laboratory, Tulane University School of Medicine, New Orleans.* The addition of 20 gm. of gelatin per day for 10 days to a wheat diet providing about 40 gm. of protein and 6 mg. of nicotinic acid daily (J. Biol. Chem., **167**: 293, 1947) has no effect upon the urinary excretion of N′-methylnicotinamide (N′-Me) or of 4-pyridoxic acid. There is a decrease in excretion of tryptophane and indoleacetic acid-like compounds related to tryptophane. The simultaneous addition of 200 mg. of dl-tryptophane with this gelatin results in an increased excretion of N′-Me and tryptophane with no change in 4-pyridoxic acid. When 5 mg. of pyridoxine is also incorporated into this diet, the excretion of N′-Me is decreased and that of 4-pyridoxic acid increased. The addition of 20 gm. of glycine per day for 10 days to the original wheat diet does not affect the excretion of N′-Me or 4-pyridoxic acid, but does lead to an increase in tryptophane excretion. The glycine ingestion results in larger increases in urinary nitrogen than does the gelatin. Subjects on a corn diet, in which three-fourths of the unenriched flour of the wheat diet is replaced by corn meal and grits, show somewhat similar responses when pyridoxine, gelatin, or gelatin and tryptophane are added.

The double strain phenomenon: Biological basis of the nutritional effect in natural resistance to infection. HOWARD A. SCHNEIDER. *The Rockefeller Institute for Medical Research, New York, N. Y.* The resistance of mice to infection with *Salmonella typhimurium*, arranged by either genetic or nutritional means, has been found to be dependent upon the presence of some avirulent cells in the heterogeneous infecting inoculum. This fact has led to the separate use of avirulent and virulent cultures in the infection procedure. By controlling the dose and the time interval between the separate injection of avirulent and virulent cultures, the effect of diet on survivorship has been analyzed and an optimum infection procedure selected at which the dietary effect is maximal. The assay of foodstuffs for nutrients capable of influencing an infection has thus become a practicable matter. Such diet-infection experiments are now performed by injecting the mice intraperitoneally with 10^8 viable cells of an avirulent strain of *S. typhimurium* and after 24 hours re-injecting the mice with 10^3 viable cells of a homologous virulent strain. The results of fractionation of foodstuffs for resistance and susceptibility factors will be presented.

Effect of phytate and other food ingredients on the absorption of radioactive iron. LEON M. SHARPE (by invitation), ROBERT S. HARRIS, WENDELL C. PEACOCK (by invitation), and RICHARD C. COOKE (by invitation). *Departments of Food Technology and Physics, The Massachusetts Institute of Technology and the Walter E. Fernald State School.* Fifteen boys, 12 to 17 years old, living in an institution, were fed breakfasts of the following test meals: I, 200 ml. water; II, 200 ml. milk; III, 200 ml. milk plus 234 mg. phytic acid as the sodium

salt; IV, 200 ml. milk, plus 234 mg. phytic acid as in rolled oats (285 gms.); V, 200 ml. milk, plus 285 gms. rolled oats, 150 ml. tomato juice, 75 gms. egg omelet and 56 gm. white bread. Total iron content of all meals was equalized with V at 8.31 mg. using $FeCl_3$. Ascorbic acid (32 mg.) added to reduce all iron to ferrous form. I and II contained no phytate. Fe^{55} and Fe^{59} were used alternately. No food was eaten before or until five hours after the test meals. Radioactive iron absorption was calculated from activity of hemoglobin, plasma volume and hematocrits. Maximum radioactivity of the hemoglobin was reached within 10 days after feeding. Average percentage of ingested iron appearing in the hemoglobin was: I, 12.4; II, 7.7; III, 0.8; IV, 4.8 and V 2.5. Sodium phytate markedly affected iron absorption and is five times more reactive than that in oatmeal. Milk and milk plus oatmeal decreased iron absorption by ⅓rd and ⅔rd, respectively. Thus phytate-free milk interfered with iron absorption nearly as much as phytate-rich oatmeal.

Physical factors influencing dental caries in the cotton rat. J. KNOX SMITH, E. POTTS ANDERSON, MARIE ZEPPLIN, C. A. ELVEHJEM and PAUL H. PHILLIPS. *Department of Biochemistry, University of Wisconsin, Madison, Wisconsin.* In the production of dental caries in the cotton rat there were certain results which could not be explained in terms of the dietary constituents. Consequently the effect of the physical state of the ration was more thoroughly investigated. Since milk seemed to offer protection against dental caries even when fed along with solid cariogenic rations, diets were used which could be fed in the solid form or liquid form. Dry whole milk (Klim), dry skim milk plus butter, and a casein sucrose diet were used in the study. When any of the above diets were reconstituted with water and fed; a very great reduction in carious lesions were noted compared to the carious lesions of the control animals. The particle size of the ration was found to be important. A ration containing 67% carbohydrate (¾ dextrin, ¼ sucrose) diet was used. The dextrin in the diet was fed as coarse or floured. The floured dextrin ration resulted in over twice the amount of dental caries. Caloric restriction studies have also been made. It has been observed that a considerable reduction of carious lesions can be obtained by restricting the food intake of cotton rats.

A new dairy food. BARNETT SURE. *University of Arkansas, Fayetteville.* The dairy food consists largely of various milk solids, supplemented with dried brewers' or cultured yeasts, iron and calcium salts, and enriched with vitamins A and D. It has a protein content of 21.8%; a water content of 54%; a mineral content of 4.3%; and a fat content of about 3.5%. Twenty four albino rats have made excellent growth during an eight week period when this food served as the only sources of proteins, fat, minerals, vitamins and calories. In other words, it is a complete food. Over 200 pounds of this food have been canned to-date and the canned product has kept well over four months, being just as fresh and moist after this period as the day after canning. About 400 test meals have been now completed with children and adults ranging in ages from 2 to 75 years who have found the basic food and the food products prepared from it very palatable. The dairy food has been served as fried patties in sandwiches, as dairy loaves, as dairy balls with Spanish sauce and spaghetti, and as croquettes. The loaves and dairy balls have distinct meaty taste. Because of the low cost of the ingredients, it should be possible to make this dairy food available here and abroad at relatively low prices to the consumer.

Biological value of proteins in food yeasts. BARNETT SURE and FRANCES HOUSE (introduced by BARNETT SURE). *University of Arkansas, Fayetteville.* This work was carried out on albino rats with the Mitchell method for nitrogen balance studies. Three dried food yeasts (Anheuser-Busch) were studied: brewers' yeast, K; and cultured yeasts, G and 300. On a 5 per cent protein level, the biological values and true digestibilities were respectively, as follows: yeast K, 66.3 and 91.4; yeast G, 69.8 and 91.3; and yeast 300, 71.2 and 92.2. On a 8 per cent protein level, the biological values and true digestibilities were respectively, as follows: yeast K, 65.6 and 88.4; yeast G, 68.1 and 86.4; and yeast 300, 73.1 and 87.9.

Evaluation of adequate protein nutrition. P. SWANSON, W. W. SMITH, (by invitation), M. BRUSH, (by invitation), and H. MERRIAM, (by invitation). *The Nutrition Laboratory of the Iowa Agricultural Experiment Station, Ames.* In an attempt to define adequate nutrition with respect to protein in the albino rat, biochemical and physiological measurements that may serve as indices of protein nutrition have been made on 50 healthy stock rats. These data have been correlated with other information obtained in metabolism experiments. The quantity and quality of the dietary supply of nitrogen needed to support this state of nutrition has been studied. Groups of animals have been maintained on diets containing the single protein, lactalbumin, in concentrations ranging from one to 18 per cent of the diet by weight. The lowest quantity of lactalbumin capable of maintaining the animals in the state of nutrition characteristic of the stock controls has been determined. Studies of the dietary importance of various amino acids are in progress at the present time.

Studies in the rat of inhibitors of pteroylglutamic acid structurally related to this vitamin. M. E. SWENDSEID (by invitation), E. L. WITTLE (by invitation), G. W. MOERSCH (by invitation), O. D. BIRD and R. A. BROWN. *The Research*

Laboratories, Parke, Davis and Co., Detroit, Michigan. Several inhibitors of pteroylglutamic acid (PGA), active in bacteria, including pterins and crude PGA analogues, were administered to weanling rats to determine whether they produced symptoms characteristic of PGA deficiency, *viz.* reduction in growth rate and decrease in cellular elements of the blood. Alleviation or prevention of these symptoms by administration of PGA was taken as further evidence that the compounds were inhibitors of PGA for the animal. Data are presented on the following compounds: 2,4-diamino-6,7-diphenylpterideine; 2,4-diamino-6,7-dimethylpterideine; 2,4-diamino-6,7-di(4-amino-phenyl)pteridine; 2 amino-4-hydroxy-6,7-diphenylpterideine; N-[4-{[(2,4-diamino-6-pteridyl)-methyl]-amino}-benzoyl]-glutamic acid ("4-amino pteroylglutamic acid"); and 4-desoxypteroylglutamic acid. By means of an assay procedure based on the development of leucopenia and/or anemia within a 14-day period in the weanling rat receiving a purified diet containing succinyl-sulfathiazole supplemented with the test compound, "4-amino PGA" was found to be the most efficient PGA inhibitor, with 2,4-diamino-6,7-diphenyl pterideine also showing a considerable effect. It was found that the amount of PGA necessary to counteract increasing levels of "4-amino PGA" was not proportional to the amount of inhibitor. Some evidence was obtained, on the basis of leucocyte determinations that a compound showing inhibition of PGA at a given level will show PGA activity at a lower concentration. Under our experimental conditions the PGA analogues produced both an anemia and leucopenia whereas the pterine had a preferential effect on the leucocytes. PGA analogues decreased the apparent PGA content of the liver while pterins had no such effect. The changes in leucocyte and hemoglobin concentrations in 150–180 grams rats receiving a purified diet supplemented with "4-amino PGA" are discussed.

Influence of pteroylglutamic acid on the synthesis and action of the antipernicious anemia factor. ARNOLD D. WELCH, ROBERT W. HEINLE (by invitation), JACK A. PRITCHARD (by invitation), and HERBERT SALIS (by invitation). *Departments of Pharmacology and Medicine, Western Reserve University School of Medicine.* Swine fed purified diets containing succinylsulfathiazole and an antagonist of pteroylglutamic acid (PGA), developed leucopenia and macrocytic anemia with megaloblastic hyperplasia of the bone marrow. Remission followed the administration of either PGA or refined liver extract. Relapses developing after liver-induced remissions were refractory to liver extract, but subsequently responded to PGA. Hematologic remission was induced in one pig by the intramuscular administration of 4 daily 10 mg. doses of PGA. Four weeks later the maintained remission was unaffected by 13 additional doses of PGA, but extract prepared from liver (200 gm.) removed aseptically was hematopoietically nearly inactive in a pernicious anemic patient. After a month on the same diet, except that crude casein containing extrinsic factor replaced the purified casein, the pig was sacrificed and a second liver extract was prepared. This invoked, in the same patient, a reticulocytosis (12%) without clinical improvement or increase in erythrocyte count; extract prepared from normal pig liver caused a subsequent reticulocytosis (28%) with striking clinical improvement and a prompt increase in the number of erythrocytes. Thus, PGA can elicit a complete hematopoietic response in a pig on a purified diet free of extrinsic factor. If the synthesis of antipernicious anemia factor is involved in this response, little or no hepatic storage of the factor occurs. Administration of extrinsic factor, without added PGA, results in storage of antipernicious anemia factor, but the amount is much less than in a normal animal. The significance of these and other experiments will be discussed.

Lipid components of skin of dogs on low fat diet and dogs receiving lard. HILDA F. WIESE (by invitation) and ARILD E. HANSEN. *Dept. of Pediatrics, Univ. of Texas Medical School, Galveston.* Lipid studies have been made on the skin of 29 dogs. These included 9 newborn puppies, 9 young dogs on a low fat diet and 11 control dogs who received fresh lard in the diet. All the dogs on the fat deficient diet manifested desquamation of the skin and dry coarse hair which are characteristic for dogs receiving this diet. The appearance of the skin of the newborn puppies and the dogs receiving fresh lard as 29% of their caloric intake was that of normal healthy animals. The total fatty acids extracted from the skin of newborn puppies showed the highest iodine numbers in the three groups of dogs, while the fatty acids from the skin of dogs on the fat deficient diet were the least unsaturated of the group. There was no difference between the three groups in the average per cent of cholesterol in the skin, but the average cholesterol ester content of the newborn puppy's skin was only ⅓ of the other two groups. In addition, the desquamated portion of skin which can be peeled off readily from fat deficient animals showed a composition markedly different from the skin as a whole. Cholesterol and cholesterol ester percentages in this portion of dry skin are more than seven times those of a section of the whole skin. Also the average iodine number of the total fatty acids in the desquamated tissue falls far below that of any other tissue.

THE AMERICAN ASSOCIATION OF IMMUNOLOGISTS

THIRTY-SECOND ANNUAL MEETING

Atlantic City, New Jersey, March 15, 16, 17, 18, 19, 1948

(For possible corrections in any of the following abstracts see the next issue.)

On the purification of the rickettsia of tsutsugamushi disease (scrub typhus). CHARLES A. BAILEY, FRED H. DIERKS and JAMES E. PROFFITT (introduced by A. F. COCA). *National Naval Medical Center, Naval Medical Research Institute, Bethesda, Maryland.* A method for the preparation of a relatively pure suspension of rickettsia from the yolk sacs of embryonated hen's eggs infected with *Rickettsia orientalis* has been devised. The suspension has been used as an antigen in serological studies of scrub typhus immune sera by complement fixation tests and as a vaccine in immunity studies of scrub typhus in the white mouse.

The isolation and properties of specific antigens from variants of Shigella sonnei. EDGAR E. BAKER (by invitation), ELY PERLMAN and WALTHER F. GOEBEL. *The Rockefeller Institute for Medical Research, and the Mt. Sinai Hospital.* Variants of *Shigella Sonnei* have been described which are known as Phase I (smooth), Phase II, and Rough, and are serologically and morphologically distinct. Specific antigens have been isolated from the Phase I and II variants which are electrophoretically homogeneous. These antigens appear to be lipocarbohydrate-proteins which are toxic and highly antigenic in experimental animals. The gross chemical properties of the two antigens are similar to those obtained from the Flexner group of dysentery bacilli, yet their immunological properties are distinct and specific. The Phase I antigen contains 5.98 and 2.3 per cent nitrogen and phosphorus respectively. The Phase II antigen contains 5.5 per cent nitrogen and 3.1 per cent phosphorus. Antisera prepared by injecting groups of rabbits with minute amounts of the purified antigens agglutinate specifically the homologous microorganisms and show no serological crossing with organisms of the heterologous phase. From the results of these experiments it appears that this phase variation is associated with a change in the enzyme systems of the microorganism which govern the synthesis of the specific somatic antigen.

A critical analysis of the performance of the photronreflectometer in the measurement of serological and other turbid systems. ELLIS BOLTON (by invitation), CHARLES LEONE (by invitation) and ALAN BOYDEN (by invitation). *Bureau of Biological Research and Department of Zoology, Rutgers University.* The Libby photronreflectometer (photroner) is a simple, effective instrument for the measurement of turbid systems. Since it is used as a quantitative tool it is of importance to determine its limitations for the task of providing the quantitative data by means of which serological problems may be solved. Toward this end four photroners were used to measure; (1) inorganic precipitates, (2) a non-specifically precipitated protein suspension, (3) antigen-antibody precipitates. The results of these measurements reveal some differences in the behavior of relatively simple, homogeneous precipitates (such as those of clay suspensions) when compared with complex antigen-antibody precipitates. The difference in dielectric properties of the precipitates and the fact that the specific antigen-antibody precipitate system is a dynamic one may account for these differences in behavior of the inorganic vs. the specific precipitate system. In the latter, the changes in particle size and in the rate of size increase, occur in a system of increasing total amount of precipitate and influence each other in such a way that the net turbidity readings are proportional to the actual amounts of precipitate formed ("internal compensation"). Thus it has been found that over usual ranges of antibody concentration total turbidity (curve area) is directly proportional to the total mass of precipitate formed; photronreflectometric measurements of antigen-antibody precipitates are relatively insensitive to moderate changes in particle size; the upper limit of instrument response depends upon the suspension studied as well as the instrument used.

A study on toxins and antigens of S. dysenteriae. DANIEL A. BOROFF. *Camp Detrick, Frederick, Maryland.* Two types of toxin have been described by various investigators working with *Shigella dysenteriae.* One is associated with the somatic antigen and is soluble in diethylene glycol, while the other is an endotoxin that is precipitated from autolysates of these organisms by trichloracetic acid. Rough variants lack the somatic antigen but possess the endotoxin. Our investigation has revealed, that although the endotoxin from smooth variants induces formation of antibodies capable of protecting animals against the endotoxin of the rough variant, the latter is neither antigenic nor serologically active. Chemical analyses of the two endotoxins showed that while the nitrogen and phosphorus content of both were similar, the endotoxin of the rough variant was poorer in carbohydrate. By means of absorption experiments, the intact organisms, a diethylene glycol extract, or the trichloracetic acid precipitate from an autolysate

of the smooth variant of *S. dysenteriae* were found to induce and react with identical antibodies. Antisera to these substances were completely exhausted by absorption with any one of the above preparations. These observations are not compatible with the theory of the existence of two or more toxins and the view that the somatic antigen and the endotoxin are distinct antigenic and toxic entities. To explain the phenomena observed, the suggestion is offered that there may be only one dominant antigen in *S. dysenteriae*, consisting of a complex composed of a toxic non-antigenic protein portion and a specific carbohydrate substance that is responsible for the serological and immunological activity. Presumably, this complex may split into various proportions of protein and carbohydrate, depending upon the method of treatment, to yield products described by various investigators as distinct components of *S. dysenteriae*.

Characterization of the "enzymic" action of influenza viruses on human red cells. B. A. BRIODY (introduced by THOMAS FRANCIS, JR.). *Walter & Eliza Hall Institute, Melbourne, Australia.* Recent publications from this laboratory have shown that changes in human RBC including the removal of virus-receptors and the development of "pan-agglutinability" can be produced by filtrates of *Vibrio cholerae* and by the viruses of mumps, Newcastle disease, and influenza. Considerable evidence has been presented which indicates that these alterations of the surface character of human RBC are the functions of an enzyme in the bacterial filtrates and the virus particles. This parallelism to a standard "enzymic" action is most clearly seen in that aspect of the virus-cell reaction in which concomitantly the cell-receptor is destroyed and the virus eluted. It will be shown that two capacities of the influenza group of viruses (a) the agglutination of RBC, and (b) the elution of the virus particles from the RBC with concomitant receptor destruction, can be dissociated by appropriate treatment. The elutability of viruses is influenced in general by the same treatments and in the same direction as the activity of the soluble factor, derived from the cholera vibrio. Elutability can be destroyed by heating that is without action on the hemagglutinin titer; such heated virus can be liberated from RBC to which it is adsorbed by the cholera factor. The evidence presented is consistent with the hypothesis that a factor closely resembling the cholera factor is an integral part of the surface of these viruses and that it plays an important role in the initial virus-cell interaction.

The effect of certain enzyme inhibitors on the activity and growth of psittacosis virus. THOMAS E. BURNEY (by invitation) and ORVILLE J. GOLUB. *Camp Detrick Frederick, Maryland.* The large size of psittacosis virus suggests the presence of intrinsic enzyme systems required for its activity. Specific group inhibitors were utilized to detect the presence of an effect on —SH groups and other enzyme systems. Incubation of psittacosis virus *in vitro* with these inhibitors at 37°C. for one hour and subsequent titrations of the washed virus in embryonated eggs showed complete or almost complete inactivation with para-chloromercuribenzoate (.0001 M), hydroquinone (.001 M), proflavine (.001 M), atabrine (.001 M), and mercuric chloride (.001 M). Iodoacetamide (.0025 M) and o-iodosobenzoate (.0001 M) produced partial inactivation of the virus. Mono-iodoacetic acid, 2,4-dinitrophenol, sodium azide, sodium fluoride, potassium cyanide, PABA, and urethane produced no inactivation. Glutathione or cysteine was shown to prevent or reverse the inactivating effect of para-chloromercuribenzoate on the virus. Dependence of the virus on certain extrinsic enzyme systems of its host was suggested by reduced yields of psittacosis virus in tissue cultures which had previously been exposed for one hour to o-iodosobenzoate (.001 M) or hydroquinone (.001 M). No significant reduction in yield was observed when the remainder of the compounds listed above were tested. These results point to the importance of —SH containing enzyme systems in the activity of psittacosis virus.

Excretion of antibody in feces and urine and its absorption from the bowel. WILLIAM BURROWS. *Department of Bacteriology and Parasitology, University of Chicago, Chicago, Illinois.* Antibody is shown to be excreted in both urine and feces of guinea pigs and man immunized with cholera or typhoid vaccine, reaching agglutinin titers of 1:100 in undiluted morning urine or 24 hour specimens, and in saline extracts of feces containing 10–25 mg/ml solids before clarification. Following primary inoculation peak titers are independent of peak serum titer, appearing in feces, urine and serum in that order, suggesting that the excreted antibody is not derived from serum antibody. It is shown that serum globulin is normally excreted by these routes, and antibody titer is a function of the relative proportions of normal and immune globulin. Fecal and urinary titers decline rapidly, disappearing in 3 weeks in the guinea pig and in 9 weeks in man, but may be maintained by periodic reinoculation. Both homologous and heterologous serum antibody are excreted in the urine and feces following passive parenteral immunization. When given by intragastric inoculation, immune globulin is absorbed from the gut as demonstrated by the titration of antibody, including diphtheria antitoxin, and, in the case of heterologous serum antibody, by the presence of the foreign globulin as shown by the precipitin test, in the serum of the recipient. Peak titer in the serum of the recipient appears 10–14 days after intragastric inoculation, suggesting that the absorbed antibody is stored for

a time in tissues other than the blood. Secretion in mucus and transport by lymphocytes are suggested as possible mechanisms of passage of antibody into the bowel from the tissues.

Diphtheria toxin-antitoxin reaction in human antisera. MELVIN COHN and A. M. PAPPENHEIMER, JR. *Dept. of Bacteriology, New York University College of Medicine.* Using quantitative immunochemical methods to study the diphtheria toxin-antitoxin reaction in hyperimmune rabbit, horse and human antisera it has been shown that (1) the human system resembles that of the rabbit rather than that of the horse. (2) The quantitative precipitin curve in both human and rabbit systems can be described by the equation of Heidelberger and Kendall. (3) There are no significant differences in the quantitative precipitin curves using antisera obtained from persons showing tuberculin type hypersensitivity (pseudoreactions) to diphtheria toxoid and using antisera from non-sensitive individuals. The ability of human antitoxin to give a precipitin reaction with toxin is destroyed by heating at 56°C. for 30 minutes but its protective power is only slightly impaired. The altered antitoxin present in heated antisera can be quantitatively precipitated on the rabbit toxin-antitoxin precipitate. On fractionation of human antisera by means of alcohol at low temperature, following the method of Deutsch et al., diphtheria antitoxin was found largely in the γ_2- globulin fraction.

Complement fixation with soluble antigens of Plasmodium knowlesi and Plasmodium lophurae. DAVIS, BERNARD D. (introduced by JULES FREUND). *From the Public Health Research Institute of the City of New York, Inc., N. Y. C. 9.* Soluble antigens prepared from *P. lophurae* (duck blood) and *P. knowlesi* (monkey blood) fixed complement with sera, frequently in high titer, of animals infected or immunized with the homologous parasites. The lophurae antigen also cross-reacted with anti-knowlesi monkey sera, but the knowlesi antigen did not react with anti-lophurae duck sera. The knowlesi antigen was an $(NH_4)_2SO_4$ precipitate from a hemolysate. Lophurae parasites, however, yield little antigen to hemolysis or isotonic extraction. The lophurae antigen was prepared by 10% NaCl extraction, which not only eliminates the undesirable stromata and red cell nuclei, but also yields a larger amount of antigen, and a less anticomplementary product, than the original suspension of parasites. The lophurae antigen appears to be associated with intracellular particulates; in contrast to the more truly soluble knowlesi antigen, it forms an opalescent solution, its activity is sedimented at 16,000 rpm and is destroyed by lyophilization, and it cannot be extracted from parasites treated with 0.1% formalin. It fixed complement when tested with a few human malarial sera. Much weaker reactions were obtained from a comparable precipitate from normal monkey hemolysate, and none from a comparable 10% NaCl extract of normal duck red cells.

Complement activity of sera from healthy and sick children. PAUL F. DE GARA and HENRY P. GOLDBERG (by invitation). *New York Hospital and the Department of Pediatrics, Cornell University Medical College, New York City.* Complement was titrated by the "50 per cent Hemolysis Method" in sera from 107 children (age range 3 days to 15 years) who were well at the time of the test and for a preceding four-week period. Titers ranging from 0.0040 to 0.0069 ml. were found in 83.5 per cent, higher titers in 6.3 per cent, and lower titers in 10.2 per cent. The mean titer was 0.0053 ml. + 0.00115. There was no relationship between complement activity and age, sex or season at which the specimen was obtained. Low complement titers were found in 20 per cent of children who were sick at the time of the test (upper respiratory infections, pneumonia, meningitis, rheumatic fever, nephritis and other diseases).

Attempt to produce protection against mosquitoes by active immunization. I. N. DUBIN, J. D. REESE (by invitation) and LOIS A. SEAMANS (by invitation). *Div. of Pathology and Bacteriology, Univ. of Tennessee College of Medicine, and Dept. of Health and Safety, Tennessee Valley Authority.* It has been suggested that the tolerance which some persons acquire to the bites of blood-sucking insects is the result of a true immunity. Trager demonstrated that ticks incited an acquired immunity after feeding on experimental animals. As yet, no true anti-insect immunity has been proven in regard to adult mosquitoes. The present experiment was undertaken to see whether rabbits could be protected against mosquitoes by active immunization. Antigens consisted of: (1) 1:100 aqueous suspension of ground-up bodies of female *Anopheles quadrimaculatus*, (2) Seitz filtrate of extract of above suspension by repeated freezing and thawing. The antigens were mixed with mineral oil and lanolin. Normal and test animals were exposed to hungry mosquitoes under standard conditions. The mosquitoes fed equally well on both control and test groups, and were not inhibited in their biting. Moreover, the group of rabbits inoculated with the crude suspension of mosquitoes became sensitized to mosquito bites in contrast to normal non-sensitive rabbits. The cutaneous reactions consisted of large indurated red papules at the site of the bites; they began in about 30 minutes and reached their maximum size at about 5 hours. Attempts to desensitize the animals with the two antigens were unsuccessful. Passive transfer of the sensitivity to normal animals was also tried, but without success. Thus, the course of vaccination afforded no protection to the animals against mosquitoes, but instead produced hyper-

sensitivity to mosquito bites in previously non-sensitive animals.

Studies in hypersensitivity in simian malaria. I. N. DUBIN. *Div. of Pathology and Bacteriology, Univ. of Tennessee College of Medicine.* Antigens prepared from *Plasmodium knowlesi* and *P. gallinaceum* were inoculated intracutaneously into monkeys in the acute and chronic stages of infection with *P. knowlesi* or *P. cynomolgi*. The antigens prepared from *P. knowlesi* consisted of: (1) aqueous suspensions of dried parasites (2) aqueous extract of dried parasites at room temperature for 1 hour (complement-fixation antigen used by Dulaney and Stratman-Thomas) (3) aqueous extract of dried parasites in refrigerator for 24 hours (4) aqueous extract of dried parasites by repeated freezing and thawing (5) solution of wet parasite mass in N/10 NaOH and restoration of pH to 8.0 by H_3PO_4. The antigens prepared from *P. gallinaceum* consisted of: (1) aqueous suspensions of "Malarial Antigen (Dried)" Lederle (2) aqueous extract of above dried antigen at room temperature for 1 hour. As controls, preparations were made from hemolyzed blood of normal monkeys and chickens. The results of the tests were negative; no significant cutaneous reactions were found. An attempt was made to sensitize monkeys to malarial antigen by the use of adjuvants such as a mixture of lanolin, mineral oil, dead tubercle bacilli and dried malarial antigen (*P. knowlesi*); again no cutaneous hypersensitivity was demonstrated. Macrophage sensitivity was sought for in tissue cultures by suspending macrophages from normal monkeys and monkeys chronically infected with *P. knowlesi* in plasma containing dried malarial antigen (*P. knowlesi*); no evidence of macrophage sensitivity in the diseased monkeys was elicited. As judged by these experiments, there is no significant development of hypersensitivity in simian malaria, either accompanying infection or following artificial sensitization.

Studies on Donovania granulomatis. 2. Preparation of culture antigens and immunologic tests. ANNA DEAN DULANEY, KODA GUO and HENRY PACKER. *Division of Pathology and Bacteriology, and Preventive Medicine, University of Tennessee College of Medicine, Memphis.* Four antigens were prepared from *Donovania granulomatis* grown on Locke-embryonic yolk-medium. These consisted of bacterial suspension, filtrate, boiled filtrate, and dialyzed filtrate antigens. Of these, the bacterial suspension and boiled filtrate antigens proved most satisfactory, and were employed in complement fixation tests with sera from 88 individuals. These sera were obtained from 24 patients with granuloma inguinale established by positive smears, 28 patients with early syphilis, 5 patients with lymphogranuloma venereum, 11 patients with miscellaneous genital lesions, and 20 healthy individuals with no evidence of venereal infection. The highest order of sensitivity and specificity was obtained with the boiled filtrate antigen. Eighty-three per cent of 24 patients with granuloma inguinale gave positive reactions with this antigen. One weak (1 +) non-specific reaction was obtained in the syphilis group with this antigen. In the same group of syphilitics, 6 positive tests, mostly weak in character, were obtained with the bacterial suspension antigen, as well as 5 with the control antigen. No positive tests were obtained with either the boiled filtrate or bacterial suspension antigens in the patients with lymphogranuloma, miscellaneous genital lesions, and healthy controls. Skin tests were also performed with these two antigens but the results were inconclusive. The bacterial suspension appeared to be a better skin test reagent than the boiled filtrate antigen.

Studies on inhibition of the agents of murine and feline pneumonitis by penicillin. MONROE D. EATON. *Department of Bacteriology and Immunology, Harvard Medical School.* Infections of the yolk sac caused by the agent of mouse pneumonitis were sterilized by penicillin in doses of 100 units given 24 hours after infection. For a similar complete virucidal effect on the cat pneumonitis virus, 5,000 units given three times at 2, 72, and 144 hours after infection were necessary. When treatment was delayed the dose of penicillin required increased with the time after inoculation at which it was administered, but inhibitory and virucidal effects on the mouse pneumonitis virus were demonstrable with 5,000 units given 4 to 5 days after inoculation. In vitro virucidal effects by crystalline penicillin G at a concentration of 2,000 units per cubic centimeter were not demonstrable. Impurities of unknown nature in crude penicillin were virucidal in vitro for the mouse pneumonitis virus.

Serological studies on infectious mononucleosis and other conditions with human erythrocytes modified by Newcastle disease virus. ALFRED S. EVANS (by invitation) and EDWARD C. CURNEN. *Section of Preventive Medicine, Yale University School of Medicine, New Haven, Conn.* Serological reactions with human erythrocytes modified by adsorption and elution of Newcastle disease virus (NDV) were studied to investigate the possible relationship of this virus to the causative agent of infectious mononucleosis suggested by Burnet and Anderson. Serum from 5 of 23 patients with infectious mononucleosis agglutinated such NDV-treated cells in higher titre than serum fron normal persons or from patients with non-infectious diseases. Similarly high titres were also obtained with serum from 13 of 101 patients with infections other than infectious mononucleosis. Serum from rabbits immunized against Newcastle disease virus or against any of three other immunologically unre-

lated viruses agglutinated NDV-treated cells in higher titre than normal rabbit serum. Human serum and rabbit antiserum against viruses other than Newcastle disease virus which agglutinated NDV-treated human red cells in high titre did not specifically inhibit the agglutination of chicken red cells by Newcastle disease virus. The capacity of both human and immune rabbit sera to agglutinate NDV-treated human erythrocytes appeared to be unrelated either to the heterophile antibody of infectious mononucleosis or to the hypothetical agent of that disease.

The long persistence of Rickettsiae orientalis in the blood and tissues of infected animals. JOHN P. FOX (introduced by PETER K. OLITSKY). *From the Laboratories of the International Health Division, Rockefeller Foundation, New York.* Mice and eastern cotton rats, infected with *R. orientalis* (agent of tsutsugamushi disease) and surviving because they were treated with thionine dyes or because they had been infected by the subcutaneous route, were sacrificed at various intervals after infection. Blood and tissues were tested for infectivity. Parallel studies were made of serum-antibody (complement fixing) and of resistance to a certainly lethal challenge inoculum of homologous rickettsiae. The observations on cotton rats were made for 269 days and those on mice for 610 days. In both animal species, the titer of serum-antibody soon rose to a significant level and remained constant at that level during the entire period of observation. Mice, challenged at intervals, consistently resisted the challenge infection. In spite of the evident solid immunity, it was possible to infect passage mice with one or more materials (whole blood or suspensions of brain, liver or kidney) taken from animals sacrificed at any time during the period of observation. Although kidney was the most reliable source of infectivity, whole blood was shown to be infectious up to the 102nd day in cotton rats and to the 610th day in mice. Interest attaches to the findings with blood, not only because of the simultaneous demonstration of infectivity and antibody in many single specimens, but also because of their significance to the problem of the rodent reservoir in tsutsugamushi disease.

Mode of action of thionine dyes in combatting experimental rickettsial infections of mice. JOHN P. FOX and OSLER L. PETERSON (introduced by PETER K. OLITSKY). *From the Laboratories of the International Health Division, Rockefeller Foundation, New York.* A previous report (J. Exper. Med. **85**: 543, 1947) contains an account of the remarkable activity of methylene blue and toluidine blue in combatting experimental scrub typhus in mice. The present report deals with experiments demonstrating a similar activity of these dyes against infections with murine typhus rickettsiae, and also with observations bearing on the mechanism of this antirickettsial effect. The localization and multiplication of rickettsiae in treated and untreated animals has been studied under varying conditions of dye administration, and the antirickettsial activity of a series of chemically related compounds has been determined. From these studies, it is suggested that the mechanism of antirickettsial activity is independent of the *in vitro* rickettsiacidal activity of the dyes and only partly dependent upon the irregular rickettsiostatic effect observed *in vivo*. It seems possible that the thionine dyes also operate, by virtue of their ability to mediate intracellular oxidation, to spare the cells from the usual harmful effects produced by rickettsiae. The survey of related compounds further indicates that antirickettsial activity requires some degree of structural specificity.

Studies on the mechanism of action of furacin (5-nitro-2-furaldehyde semicarbazone). MORRIS N. GREEN (introduced by STUART MUDD). *Department of Bacteriology, School of Medicine University of Pennsylvania, Philadelphia, Pa.* The inhibitory action of furacin on the growth of *Escherichia coli* was antagonised by the amino acids, L-arginine, L-lysine, DL-phenylalanine, L-glutamic acid, DL-isoleucine and vitamins of the B complex, notably thiamin, nicotinamide, pyridoxine and pantothenate. Experiments with the Warburg respirometer showed that the aerobic dissimilation of glucose was inhibited 45% by furacin. In a similar fashion the anaerobic dissimilation of glucose and pyruvate was inhibited 35% and 70% respectively. The glucose dehydrogenase system of *Escherichia coli* was also inhibited by furacin. These results offer some explanation for the antagonisms exerted by nicotinamide and thiamin, since these vitamins are related to the coenzymes involved in the dissimilation of glucose and pyruvate. Lipmann's observations may indicate a similar relation for pantothenate in the dissimilation of pyruvate. Both tryptophanase activity and arginine decarboxylase, enzymes containing pyridoxal phosphate, were inhibited by furacin 70% and 45%, respectively. Since enzymes containing pyridoxal phosphate as coenzymes are inactivated by carbonyl reagents, the possibility is suggested that furacin may be reduced by bacteria with the formation of semicarbazide.

Studies on Donovaina granulomatis. I. Locke-embryonic yolk medium for cultivation of the organism. KODA GUO, ANNA DEAN DULANEY, HENRY PACKER. *Division of Pathology and Bacteriology and Preventive Medicine, University of Tennessee College of Medicine, Memphis.* One of the strains of *Donovania granulomatis* obtained from Dr. Katherine Anderson was successfully cultivated in Locke-embryonic yolk-medium. Equal parts of yolk and Locke solution were mixed

and coagulated in slant by steam. A small amount of liquid which gave a neutral reaction collected at the bottom of the slant. Following inoculation of the Locke-yolk medium with *Donovania granulomatis* from the yolk of fertile eggs sufficient Locke's solution was added to almost cover the slant. The heaviest growth of organisms occurred in the lower levels of the Locke's solution near the slanted egg-material. The morphology of these organisms appeared identical to that observed in organisms cultured in parallel series in fertile eggs. Curved bacillary forms with bipolar granules predominated. "Safety pin" forms resulting from bulging of the central portion were also seen. Examination by darkfield illumination revealed a highly illuminated outer thin cell with markedly refractive bipolar granules. True capsules were not observed. The addition of human serum to the Locke's solution produced chain forms made up of 5 to 8 bacilli, as well as bizarre pleomorphic forms. Upon this simple medium *Donovania granulomatis* has been carried for 34 generations without variation in morphology, and such cultures have been used in the preparation of antigens which gave highly specific and sensitive complement fixation reactions.

Toxicity for paramecia of sera from cancerous and non-cancerous persons. JAMES A. HARRISON and (by invitation) MACHTELD E. SANO, ELIZABETH H. FOWLER, ROBERT H. SHELLHAMER and CAROL A. BOCHER. *From the Department of Biology, Temple University, and the Temple University School of Medicine and Hospital.* The aim of this work was to verify the report of Roskin (Am. Rev. Soviet Med., Dec., 1946) that the presence of cancers in human beings is accompanied by the occurrence of serum substances which are toxic for paramecia. Eighty sera (from 79 persons) in final dilutions of 1–20, 1–30 and 1–40 were tested with *Paramecium aurelia* on the first, second, third and eighth days after blood collection with and without the addition of fresh guinea pig serum in final dilution of 1–20. Toxicity was judged by the number of survivors from 15 paramecia after 24 hours in the presence of each serum dilution. This series contained sera from 28 persons with malignant neoplastic disturbances and from 51 persons, including 19 hospitalized individuals, with no history of cancer. Among the 28 sera from cancerous persons 8 were definitely toxic for paramecia, 6 were possibly toxic and 14 were definitely non-toxic. Among the 52 sera from non-cancerous persons 13 were definitely toxic, 10 were possibly toxic and 29 were definitely non-toxic. The toxic fraction in these sera was destroyed rapidly at a temperature of 55°C. and slowly at 4°C. The addition of fresh complement to heated of cold-stored sera did not, with a single exception, restore or enhance toxicity. Sixty duplicate and triplicate runs on 30 of these sera indicated that the determinations of toxicity were quite reliable. Essentially similar results were obtained when sera from 29 additonal cancerous and non-cancerous persons were tested with several other species of ciliates. It appears then that while human serum frequently contains fractions sharply toxic for paramecia the toxicity is not closely related to the occurrence of cancer.

Effect of environmental temperature upon sulfadiazine therapy, body temperature and oxygen consumption in pneumococcus infection. JUNGE, J. MCBROOM (by invitation) and S. M. ROSENTHAL. *Division of Physiology, National Institute of Health, Bethesda, Maryland.* The survival from type I pneumococcus infection in mice under sulfadiazine therapy is significantly influenced by environmental temperature. The optimum is in the neighborhood of 26°C. to 31°C., while markedly lowered therapeutic responses are observed at 18°C. Lowered responses also appear at 37°C. No rise in body temperature was found in mice during the course of a fatal pneumococcus septicemia. Upon placing infected animals under slight stress, by exposing them to 18°C., it was found that the oxygen consumption does not exceed levels attained by normal mice and under these conditions a progressive fall in body temperature occurs. This indicates that the fall in temperature is due to inadequate heat production rather than excessive heat loss. The evidence suggests that as a result of the infection certain mechanisms which, either directly or indirectly, control the metabolic processes are damaged to the extent that the metabolic capacity of the animal is impaired. The nature and site of this damage remains to be established.

Immunochemical estimation of albumin and gamma globulin in normal and pathological cerebrospinal fluid. ELVIN A. KABAT, MURRAY GLUSMAN (by invitation) and VESTA KNAUB (by invitation). *Departments of Neurology and Bacteriology, College of Physicians and Surgeons, Columbia University and the Neurological Institute of New York.* Antisera to crystalline human serum albumin and to purified human gamma globulin are prepared in rabbits and absorbed with heterologous antigen. The antisera are calibrated by addition of increasing known amounts of antigen to a constant volume of antiserum. After one hour at 37°C. and 48 hours in the refrigerator, the precipitates are centrifuged off, washed twice in the cold with saline and analyzed for nitrogen. Total N precipitated is plotted against quantity of antigen added. The albumin and gamma globulin in cerebrospinal fluid is determined by adding an appropriate dilution of cerebrospinal fluid to the albumin and globulin antisera such that the total N precipitated will fall on the calibration curve, measuring the total N in the washed precipitates

and reading off the corresponding albumin and globulin contents from the curves. Analyses are valid only in the region of antibody excess. In ten healthy individuals cerebrospinal fluid albumin and gamma globulin ranged from 11 to 19 and from 1.7 to 3.8 mg. per 100 ml. respectively. Fifteen of 16 patients with neurosyphilis showed marked increases in cerebrospinal fluid gamma globulin ranging from 5.6 to 116 mg. per 100 ml. The lowest values occurred in patients who had responded favorably to therapy. Eight of 14 cases of multiple sclerosis also showed significant increases in gamma globulin ranging from 5.6 to 13 mg. per 100 ml.

The inactivation of influenza virus by mercurials and reactivation by sodium thioglycolate and BAL. MORTON KLEIN (by invitation), J. H. BREWER (by invitation), J. E. PEREZ (by invitation) and BEATRICE DAY (by invitation). *Department of Bacteriology, School of Medicine, University of Pennsylvania, Philadelphia, Pa. and Hynson, Westcott and Dunning, Baltimore, Md.* The Lee strain of influenza B virus was inactivated after 5 minutes contact at room temperature with 1:4,000 bichloride of mercury, 1:1,000 metaphen, 1:3,000 phenyl mercuric nitrate, 1:100 mercurochrome and 1:1,000 merodicein. A 1:2,000 solution of merthiolate did not inactivate the virus. Inactivation was determined by loss of infectivity of the virus for 10–14 gram Swiss mice following intranasal instillation. Merodicein possessed some prophylactic activity when instilled intranasally into mice one hour before the virus. A polyvalent bacteriophage active against *Staphylococcus albus* was also inactivated by 1:10,000 bichloride of mercury. Inactivation of the PR8 strain of influenza A virus and the bacteriophage by the bichloride of mercury could be reversed *in vitro* by a 1:50 aqueous solution of sodium thioglycolate or a 1:300 aqueous solution of BAL. Reactivation of the virus was determined by mouse infectivity tests. Reactivation of the bacteriophage was determined by agar plaque counts.

The reactivation in vivo of influenza A virus by BAL. MORTON KLEIN (by invitation) and ENRIQUE J. PEREZ (by invitation) (introduced by STUART MUDD). *Department of Bacteriology, School of Medicine, University of Pennsylvania, Philadelphia, Pa.* Influennza A virus inactivated by bichloride of mercury can be reactivated both *in vitro* and *in vivo* with BAL (2,3,dimercaptopropanol). In the present work we have utilized the *in vivo* reactivation technique to determine whether the adsorption of influenza virus onto the host cells will remove it from the direct action of a chemical agent. A 1:15,000 dilution of bichloride of mercury will completely inactivate influenza virus, though the ability of the virus to become adsorbed onto cells, as determined by chick red cell agglutination, is but slightly affected. If 0.1 ml. of a 1:200 aqueous dilution of BAL is given intramuscularly to 10–14 gram Swiss mice 5–15 minutes before the intranasal instillation of bichloride of mercury inactivated virus, the BAL can reactivate the virus and the mice will die of infection with influenza virus. However, when the inactivated virus is given intranasally, followed in 30 minutes by the intramuscular injection of BAL, no reactivation occurs. Pneumococci, on the other hand, inactivated with bichloride of mercury and injected intraperitoneally into mice, can be reactivated when the BAL is given intramuscularly either before or after the inactivated bacteria. The results suggest that though a chemical agent is active *in vivo* and has a direct action on the virus, the adsorption of the virus onto the host cells will protect it from the action of the compound.

Effect of salt concentration on the agglutination of red cells by influenza virus and antibody. FRANCIS C. LOWELL, M.D. and MARGUERITE BUCKINGHAM (by invitation). *Evans Memorial Hospital, 65 E. Newton St., Boston, Mass.* The influence of various salt concentrations in the presence of 5 per cent glucose, on agglutination of red cells by influenza A virus (PR 8) was studied. No visible agglutination occurred in concentrations of 0.030M NaCl or less and at a concentration of approximately 0.003M, union of virus and red cells was inhibited. Virus was eluted when combination of virus and red cells was permitted to take place and when the electrolyte was then removed by washing in a salt-free solution. Concentrations of salt as high as 0.583M failed to cause noticeable inhibition of agglutination of red cells by virus although such salt concentrations completely inhibited agglutination of red cells by antiserum. For comparative purposes, agglutination of red cells by antibody was likewise studied with respect to the effect of various salt concentrations. The results were similar to those which have been obtained by others and differed from those obtained with influenza virus in that interference with agglutination was much less marked with low and much more marked with high salt concentrations.

The effect of MG^{++} on the complementary and coagulative activities of blood serum.* FRANK MALTANER. *From the Division of Laboratories and Research, New York State Department of Health, Albany.* J. O. DE ALMEIDA (by invitation). *From The School of Medicine, University of Sao Paulo, Sao Paulo, Brazil.* Data presented previously suggest that the enhancing effect of Mg^{++} on the hemolytic and clotting activities of serum is dependent on and secondary to that of Ca^{++}. In studies of Mg^{++} treated complement in the quantitative complement-fixation test for syphilis the enhancing action of Mg^{++} was neutralized by the inactivated

human test serum. The result is an increase in the anticomplementary reaction with serum alone essentially proportional to the increase in hemolytic activity of the complement resulting from treatment with Mg^{++}. Thus, the titer of sera expressed as the ratio of the reaction obtained with serum plus antigen to that with serum alone is not altered by the use of Mg^{++} treated complement. Failure to take account of the neutralizing effect of test serum for Mg^{++} treated complement will lead to errors in the evaluation of titer. In parallel tests of the accelerating effect of Mg^{++} on the clotting activity of complement treated with Ca^{++} and cephalin, inactivated human sera neutralized also the clotting effect of Mg^{++}.

On the kinetics of hemolysis by antibody and complement. MANFRED M. MAYER and CHARLES C. CROFT (by invitation). *Dept. Bacteriology, Johns Hopkins School of Hygiene and Public Health.* Quantitative studies of the kinetics of immune lysis of sheep erythrocytes have yielded evidence indicating that the action of hemolytic antibody resembles that of an enzyme, while complement appears to play the role of a co-factor which is used up in the process. It is thought that complement may furnish the energy which enables antibody to perform its hemolytic function.

Reactivation of overneutralized influenza virus in chicken embryos. A. P. MCKEE (by invitation) and WM. M. HALE. *Univ. of Iowa, Dept. of Bacteriology, Iowa City, Iowa.* The addition of concentrated, heat-inactivated homologous virus to influenza virus type A (PR8), that was overneutralized from 50 to 10,000 times demonstrated that the neutralized virus could be reactivated. This same strain of virus could be recovered from either convalescent or artificially actively immunized mice for a longer period of time by the use of the concentrated, inactive virus than without it. Chicken embryos were used in all cases to detect the active virus. The virus used to affect reactivation was concentrated either by adsorption on and elution from chicken red cells or by evaporation after dialysis. Earlier studies concerning the effect of heat (57°C.) on the hemagglutination and antibody-binding activities of the virus were extended. As a result of heating, the hemagglutination activity of both type A (PR8) and type B (Lee) influenza virus was progressively decreased when the virus had been concentrated by the red cell method. The hemagglutinating activity of type B (Lee) was not materially affected, however, if it was concentrated by evaporation. The antibody-binding activity of type A (PR8) virus but not type B (Lee) virus was diminished by the same heat treatment regardless of the method used to concentrate the virus.

Adaptation of poliomyelitis strains to rodents with a word on nomenclature. JOSEPH L. MELNICK and ROBERT WARD (by invitation). *Section of Preventive Medicine, Yale University School of Medicine, New Haven, Conn.* This report is concerned with the adaptation of two additional strains of poliomyelitis virus to rodents: the Yale SK strain isolated in 1938 from the stools of a non-paralytic poliomyelitis patient in New Haven, and the Ph strain, isolated in 1943 from a fatal case in the Middle East. The latter strain was established in rodents with material derived from the spinal cord as well as material derived from the colon contents of this patient. Primary transfers from monkeys were made directly to mice and to cotton rats. Several serial transfers were necessary to produce the disease regularly in rodents and to standardize the titration end-point. The disease in mice is similar to that produced by Armstrong's Lansing strain and is characterized by flaccid paralysis of one or more limbs. Both strains remain infectious for monkeys: infected mouse as well as cotton rat CNS has been shown to have a hundred to thousandfold higher titer in monkeys than in rodents. The disease produced in monkeys with the rodent-adapted strain is a replica of the disease produced by the original parent strain. After adaptation in rodents, immunological relationships with the parent strains are retained. Both strains cross immunologically with each other, and with the Lansing strain, but are immunologically distinct from two other "non-adaptable" strains, the 1944 North Carolina strain and the 1945 Philippine Islands strain. Although ready means of designating strains immunologically as Poliomyelitis A, B, or C are not yet available, certain aspects concerning nomenclature and classification will be discussed.

The production of anti-rabbit hemolysi (rabbit-erythrolysin) in sheep, and its value for complement fixation tests. A. PACKCHANIAN. *University of Texas, School of Medicine, Galveston, Texas.* Anti-rabbit hemolysin was produced in sheep by injecting the sheep intravenously and subcutaneously with washed rabbit red blood cells. It was noted that the agglutination titer was much higher than the hemolytic titer. This anti-rabbit hemolysin, plus 5% washed rabbit cells, was used in complement fixation tests. The results were in every case identical with the results obtained from the usual hemolytic system; i.e., positive serum gave positive results, and negative serum, negative results. Other work is in progress to increase the hemolytic titer of anti-rabbit hemolysin. If the titer can be raised considerably, this new hemolysin will be practical for complement fixation tests in places—such as small laboratories—where sheep cannot be kept for a continuously fresh supply of red blood cells.

The zone of activity of antibodies as determined by the use of radioactive tracers. DAVID PRESS-

MAN and GEOFFREY KEIGHLEY (introduced by A. F. COCA). *Gates and Crellin Laboratories of Chemistry and the William G. Kerckhoff Laboratories of the Biological Science, California Institute of Technology, Pasadena, Calif.* We have found that a method of locating the zone of activity *in vivo* of antibodies prepared against animal tissues is to iodinate the antiserum with iodine containing radioactive iodine in tracer concentrations and determine the disposition of the radioactivity. We found that antiovalbumin serum can be iodinated without destroying its ability to precipitate specifically with ovalbumin and that antibody thus specifically precipitated contains iodine. Anti-rat kidney serum prepared according to the method of Smadel and similarly iodinated, upon inoculation into rats, localized in the kidney; while iodinated antiovalbumin serum or normal serum showed no such localization. This demonstrates that the zone of activity of the antikidney serum is the kidney.

Parallel use of the "direct" and "indirect" complement-fixation tests. CHRISTINE E. RICE. *Division of Animal Pathology, Science Service, Dominion Department of Agriculture, Animal Diseases Research Institute, Hull, Quebec, Canada.* The wider application of complement-fixation in serologic surveys of avian and mammalian populations has been hindred by the fact that convalescent sera of certain species fail to exhibit complement-fixation with homologous antigen although the presence of antibody can be detected by other methods. Non-complement-fixing sera of some of these species however display inhibitive properties when mixed in suitable proportions with convalescent sera from another species which fix complement with the same antigen. Methods of titrating the degree of inhibitive activity in such sera have been developed which furnish titers in relatively good aggrement with those determined by other serologic methods. This method has for convenience been termed the "indirect" complement-fixation test to distinguish it from the usual or "direct" complement-fixation test. The results obtained by using the two methods in parallel in a number of bacterial and viral systems will be considered.

Further studies on the production in vitro from bacteria of substances resembling "natural" agglutinins and precipitins. EDWARD C. ROSENOW. *From the Rare Metals Institute of the California Institute of Technology, Pasadena, California, and the Longview Hospital, Cincinnati, Ohio.* Suspensions, extracts and filtrates of varying strength of different species of bacteria in isotonic NaCl solution were subjected to varying degrees of heat and varying concentrations of different oxidizing agents in the acid, neutral and alkaline range of pH. The effects of removal of polysaccharides on agglutinin and precipitin production, of filtering, dialyzing and distilling on boiling and of adsorption with homologous and heterologous bacteria and of adsorbing agents and of eluting of adsorbed antibody were determined by methods usually employed. Specific types of streptococci were isolated in studies of various diseases from the end-point of growth in serial dilution cultures of dextrose brain broth and their specificity was maintained in dense suspensions of glycerol two parts and saturated NaCl solution one part. Agglutinin and precipitin titers of solutions of artificial antibody against suspensions and extracts of the respective bacteria were determined at 48°C. for 18 hours. Specific agglutinins and precipitins were produced usually in very high titer in the acid range of pH from pneumococci and streptococci, from staphylococcus aureus, E. coli, B. mucosus, and B. subtilis roughly proportional to the polysaccharide, to the density of suspensions, extracts and filtrates, to the temperature and to the concentration or activity of the oxidizing agent. Of the oxidizing agents used, O_2 under 2,000 lbs. pressure, hydrogen peroxide, sodium perborate, calcium dioxide, manganese dioxide, potassium chlorate and permanganate, hydrogen peroxide proved best. Thermal agglutinins proved filtrable, dialyzable, and distillable, but after conjugation onto globulin they were no longer dialyzable or distillable and specific titer increased.

A poliomyelitis-like agent in hamsters inoculated with Lansing virus. F. K. SANDERS (introduced by JOSEPH E. SMADEL). *Department of Virus and Rickettsial Diseases, Army Medical Department Research and Graduate School, Army Medical Center, Washington 12, D. C.* In attempts to obtain concentrations of Lansing poliomyelitis virus greater than those in infected mouse brain, intracerebral passages were made in adult (40–60 grams) or in suckling (5–8 grams) hamsters. The virus was carried for 24 serial passages in the brains of adult hamsters. The titer of the initial virus, 315th mouse passage, was 10^{-3} and the average titer during the 24 passages in adults was 10^{-4}. Virus from the 10th and 19th passages in adult hamsters (A line) were transferred to suckling hamsters (S-1 and S-2 lines). In both instances the titer promptly rose to about 10^{-6} and remained at this level during serial passage in infants. Neutralization tests revealed that all three agents (A, S-1, S-2) were identical and cross-immunity tests with A and S-1 corroborated this. The agent multiplies more readily in infant than adult hamsters. The exact identity of the hamster virus is uncertain. Anti-Lansing monkey sera gave inconsistent results in neutralization tests with the A and S-1 viruses. Moreover, the hamster viruses were unaffected by four separate pools of normal human sera all of which neutralized the Lansing mouse strain. Both A and S-1 viruses were neutralized slightly by antisera against GD VII mouse encephalomye-

litis; the mean neutralization index in five tests was 40 with hamster virus and 290 with GD VII. It appears not impossible that our hamster virus represents a poliomyelitis-like agent of hamsters. However, no such agent was encountered in 48 consecutive intracerebral passages in hamsters of Fort Bragg fever nor was the virus recovered from the feces of normal adult hamsters. The need for criteria for differentiating various rodent strains of poliomyelitis and poliomyelitis-like viruses is evident.

The relation of tryptophane utilization to the mechanism of resistance to sulfonamides. M. G. SEVAG and EDWARD STEERS (introduced by M. G. SEVAG). *Department of Bacteriology, School of Medicine, University of Pennsylvania, Philadelphia, Pa.* Five different pairs of susceptible and corresponding resistant strains of *Staphylococcus aureus* were studied by determining: (1) The tryptophane content of the cells; and (2) the amount of tryptophane consumed during growth. The growth of the susceptible organisms is markedly inhibited by sulfathiazole, whether or not the medium contains added tryptophane. In contrast the resistant organisms show no inhibition of growth by sulfathiazole in these media. Added tryptophane, in the absence of sulfathiazole, is utilized by susceptible and resistant cells whether or not glucose is present. Added tryptophane, in the presence of sulfathiazole, is not utilized by the susceptible but is utilized by the resistant cells. Glucose counteracts to some degree this failure of the susceptible cells to utilize added tryptophane in the presence of sulfathiazole. The tryptophane content of those susceptible cells which do grow in the presence of sulfathiazole indicates that the susceptible cells are capable of *synthesizing* tryptophane. The *utilization* of the synthesized or added tryptophane for growth of the susceptible cells is blocked by sulfathiazole, but the utilization of the added or synthesized tryptophane in the resistant cells is not blocked by sulfathiazole. In the absence of added tryptophane glucose does not aid in the utilization of synthesized tryptophane by the susceptible cells. The utilization of tryptophane and glucose simultaneously is concomitant with the decrease in the inhibition of growth by sulfathiazole. The principal difference between the susceptible and resistant strains with respect to tryptophane resolves itself into the failure or ability to *utilize* the added or synthesized tryptophane in the presence of sulfathiazole.

Studies on pneumococcal enzymes involved in resistance to drugs. M. G. SEVAG and JOSEPH S. GOTS (introduced by M. G. SEVAG). *Department of Bacteriology, School of Medicine, University of Pennsylvania, Philadelphia 4, Pa.* Individual strains of three types of Pneumococcus (I, II and III) were made resistant respectively to sulfonamides, acriflavine, atabrine, optochin and propamidine. Susceptible and resistant forms in the mucoid phase were investigated for their dehydrogenase activity (Thunberg technique) in the presence of glucose, hexose-diphosphate, glycerol, lactate and ethyl alcohol. In general, the resistant strains show a definite decrease in dehydrogenase activity in one or more of these substrates. An analysis of the role of riboflavin metabolism in the resistance and action of these drugs revealed the following general findings: 1) With the exception of the sulfonamides, all the drugs inhibit methylene blue reduction by the susceptible organisms in the presence of glucose. 2) The strains resistant to growth inhibition are also resistant to the inhibition of the glucose dehydrogenase activity by the respective drugs. 3) The inhibition of methylene blue reduction by atabrine, acriflavine and propamidine is counteracted by riboflavin but not by nicotinamide and thiamine. However, the inhibition by optochin is not counteracted by riboflavin. 4) Resistant strains showing a marked decrease in dehydrogenase activity show a marked acceleration of this activity in the presence of added riboflavin but not in the presence of nicotinamide and thiamine. These findings indicate a correlation between flavoprotein metabolism and resistance to acriflavine, atabrine and propamidine. Experiments relating sulfonamides and optochin to flavoprotein metabolism are suggestive but as yet insufficient.

Immunological studies of cholera filtrates. E. SINGER, S. H. WEI and S. H. HOA (introduced by Dr. COCA). *From the Central Bacteriological Laboratory, National Institute of Health, Nanking, China.* The authors confirm Burnet's finding of a substance in cholera filtrates (beef broth containing 1% agar) possessing a destructive specific action against the epithelium of the guinea pig ileum. They find it easily heat labile (50C. for 30 min.) susceptible to acid reaction (pH 5.0 to 6.4 for 24 hrs.) and to ageing (7 to 20 days at room temperature, and to 0.3 per cent formaldehyde). It is antigenic, calling forth specifically neutralizing antibodies in injected rabbits and also in human subjects who have been vaccinated with cholera vaccines. It is not present in autolysates of the vibrios; nevertheless, it is a somatic antigen, since the injection of heat killed washed vibrios stimulates neutralizing antibodies. The authors, referring to Burnet's statement that the factor is a mucin-splitting enzyme recall that other demonstrably toxic substances have enzymatic properties. They also point out the circumstance that the specifically destructive somatic antigen of cholera presents the properties of an exotoxin rather than the usual somatic antigens.

Complement-fixation with P. knowlesi antigen for diagnosis of malaria. W. D. SUTLIFF (by invitation) and ANNA DEAN DULANEY and W. L. DAVIS (by invitation). *Kennedy Hospital and Division of Pathology and Bacteriology, University of Tennessee College of Medicine, Memphis.* The complement-fixation test for malaria, employing *P. knowlesi* antigen, was used during the year 1947-1948 to determine how often confirmation of the diagnosis could be obtained when the blood film was negative. One hundred and twenty-one tests were carried out for 103 patients. The tests were positive in 15 of 55 sera from patients without parasitemia but with histories of attacks within one year. Complement-fixation tests furnished the sole objective evidence of infection in this group. In patients with parasitemia 24 of 28 gave positive complement-fixation reactions. All tests were negative in cases with no history of malaria longer than one year before examination. Doubtful reactions occurred in 13 sera. Two of these were in patients with parasitemia and five in patients with history of recent malaria. Five were associated with other diseases which at time may simulate malaria, especially Brucellosis. It was concluded that positive complement-fixation tests were of value in the diagnosis of malaria in patients without demonstrable parasitemia.

Role of the spleen and phagocytosis in the quinine treatment of malaria. W. H. TALIAFERRO, L. G. TALIAFERRO (by invitation) and F. E. KELSEY. *University of Chicago, Chicago, Illinois.* In line with work on other infections, we have found that quinine is less effective against *Plasmodium gallinaceum* in splenectomized than in normal chickens. Experiments involving infection at various intervals after quinine indicate that quinine acts only as long as it can be found in the blood. The drug acts chiefly by inhibiting asexual reproduction of the parasites. Quinine does not increase phagocytosis by the macrophages of the spleen, liver and bone marrow as ascertained from tissue sections and smears taken at frequent intervals during treatment. Therefore, quinine stimulates neither innate nor acquired immunity because both have been shown to be associated with phagocytosis by these macrophages. Splenectomy raises the quinine blood levels following a standard intravenous dose. A study of the parasitemia in splenectomized chickens given quinine indicates that splenectomy reduces acquired, but not innate immunity. It is concluded that three independent factors operate during quinine treatment of malaria, i.e., (1) innate immunity, (2) acquired immunity and (3) the antimalarial activity of quinine. Removal of the spleen decreases acquired immunity to such an extent that any increased antimalarial activity probably resulting from an increased quinine level following splenectomy is obscured.

A turbidimetric growth assay method for the determination of the relative bactericidal activities of sera. HENRY P. TREFFERS and KATHERINE E. YAW (by invitation). *Section of Immunochemistry, Dept. of Bacteriology and Immunology, Yale University School of Medicine, New Haven.* The bactericidal action of serum plus complement on microorganisms is generally assayed from plate counts of the number of organisms which survive. The method is wasteful of time and material since generally only a few of the necessary plates give readable data, and the precision leaves much to be desired. In addition, no indication of the course of the experiment can be obtained until the plates have grown out, which does not permit replication before the complement has deteriorated substantially. In the method proposed the number of organisms surviving the action of serum and complement is determined from the amounts of growth obtained on subculture in liquid media. A suitable number of organisms is mixed with immune serum and complement and incubated. At intervals quantitative aliquots are removed and placed into broth. Similar dilutions are made from control tubes containing organisms, serum and inactivated complement. All tubes are incubated until a readable density of growth is obtained. For convenience the tubes may be heated to kill the organisms before the readings are made in the colorimeter. The relative amounts of growth obtained in the subcultures are not a function of the incubation time so long as the organisms remain in the logarithmic growth phase. In experiments with *S. dysenteriae* a linear relationship has been obtained between the relative numbers of organisms surviving and the logarithm of the amount of serum added. At present all comparisons are made against a standard serum run at the same time.

A serological relationship between the virus of encephalomyocarditis and certain strains of poliomyelitis-like viruses. JOEL WARREN (by invitation) and JOSEPH E. SMADEL. *Department of Virus and Rickettsial Diseases, Army Medical Department Research and Graduate School, Army Medical Center, Washington 12, D. C.* Previous reports have indicated that the virus of encephalomyocarditis recovered from a chimpanzee in Florida produced paralytic disease and induced unique lesions in the brains and hearts of rodents and the rhesus monkey. In addition, this small size virus was shown to be immunologically unrelated to some 20 odd viruses including the SK-New Haven and Lansing strains of poliomyelitis. Finally, either this virus or one closely related to it was found to be the cause of a mild aseptic meningitis col-

loquially known as "Three-Day Fever" among U. S. troops stationed in Manila in the winter of 1945–46. The results of additional attempts to establish the taxonomic position of the virus of encephalomyocarditis (EMC) indicate its relationship to the SK-murine and M. M. strains of poliomyelitis-like agents. Animal antisera and human convalescent sera, which neutralize the EMC agent, are also capable of inactivating SK and M. M. viruses. The similarities between the EMC, SK-murine and M. M. viruses and their possible significance will be discussed.

FEDERATION OF AMERICAN SOCIETIES FOR EXPERIMENTAL BIOLOGY

PROGRAM, ANNUAL CONVENTION, 1948

Atlantic City, N. J., March 15–19

General Information

With the growth of the Federation and the increase in its requirements for meeting rooms for simultaneous scientific sessions, only a few cities are still able to accommodate the Convention without the serious inconvenience of locating facilities in different and sometimes widely scattered hotels and auditoriums. Atlantic City, with its excellent hotel facilities and its magnificent Convention Hall, meets the Federation's needs adequately and hence we are returning to it for 1948 after an interim of only one year. We enjoy a cordial welcome from the City and its Convention Bureau and Hotels. Assistance in making arrangements has been freely given by all agencies and organizations concerned.

All of the general functions of the Federation, except Society dinners, will be centralized in the Convention Hall, located at the Boardwalk between Mississippi and Georgia Avenues. Activities there include registration, scientific sessions of all Societies, symposia, motion pictures, business meetings of all Societies except the American Institute of Nutrition, the "Mixer" and commercial exhibits. A directory of council and committee meetings and special functions will be found on page 380. Haddon Hall Hotel, located at the Boardwalk and North Carolina Ave., between Central and Steel Piers, is the official headquarters Hotel.

Sunday and Monday, March 14 and 15, will be devoted to meetings of the Councils of the Societies, the Federation Executive Committee, and of various special Committees. Scientific Sessions of the six constituent Societies will begin Tuesday, March 16, at 9:00 a.m. Business meetings of the Societies are scheduled in the program.

Registration will open at 9:00 a.m. on Monday, March 15, at the Convention Hall, and will continue to Friday afternoon. The registration fee is $3.00. An official badge will be issued at the time of registration and must be worn to secure admission to any of the meetings or other activities of the Federation. Ladies who are present as guests may register without charge at a separate registration desk in the foyer of the Convention Hall.

It is urged that registration be completed on Monday, if possible, since scientific sessions begin promptly at 9:00 a.m. Tuesday.

Programs, abstracts and tickets to various special functions will be on sale in the foyer of the Convention Hall. An information desk will also be located there.

An Informal Mixer will be held in the arena of the Auditorium on Wednesday evening, March 17, from 9:00 p.m. Refreshments will be served and an orchestra will provide music for dancing. The official badge is all that is necessary for admission.

Special Functions: A list of special functions, group dinners and meetings, and their location will be found on page 380.

Placement Service: Representatives of the Placement Service office of the Federation will be available for consultation in Room 2 of the Convention Hall. Appointments between prospective employers and applicants for positions may be made there. Applicants are requested to prepare a typed statement of vitae, including age, marital status, educational, academic and research backgrounds, references and publications, and to send a copy in advance of the meeting to Dr. M. O. Lee, Director, Placement Service, 2101 Constitution Ave., Washington 25, D. C. Additional copies should be brought to the meeting by the applicant.

Hotels. Adequate hotel accommodations are available in Atlantic City and late requests can be filled.

FEDERATION PROGRAM

Thirty-Second Annual Meeting

JOINT SESSION OF THE FEDERATION

Tuesday, March 16, 1:30 p.m.

BALLROOM, CONVENTION HALL

Maurice H. Seevers, *Presiding*

1. **Paul R. Cannon,** *Department of Pathology, University of Chicago, Chicago, Illinois*
   Tissue Protein Synthesis.
2. **Louis S. Goodman,** *Department of Pharmacology, University of Utah, School of Medicine, Salt Lake City, Utah*
   Pharmacological and Physiological Aspects of Adrenergic Blockade, with Special Reference to Dibenamine.
3. **C. A. Elvehjem,** *Department of Biochemistry, University of Wisconsin, Madison, Wisconsin*
   The Nutritional Significance of the Intestinal Flora.
4. **Robert F. Pitts,** *Department of Physiology, Syracuse University, College of Medicine, Syracuse, New York*
   The Renal Excretion of Acid.

MOTION PICTURES

Thursday, March 18, 8:00 p.m.

ROOM C, CONVENTION HALL

1. **H. M. Sweeney and Biophysics Branch Personnel,** *Aero Medical Laby., Air Material Command, Wright Field, Dayton, Ohio*
   Principles of protection against the effects of negative "G".
2. **E. H. Wood, G. E. Montgomery, Jr.** (*by invitation*) **and J. E. Geraci** (*by invitation*), *Section on Physiology of the Mayo Clinic and Foundation, Rochester, Minnesota*
   A technique for obtaining multiple arterial blood samples applied to the study of cyanosis in man.
3. **J. R. Whittier** (*introduced by F. A. Mettler*), *Department of Neurology, Columbia University, College of Physicians and Surgeons*
   Rhesus hyperkinesia by subthalamic lesion.
4. **G. H. Algire and F. Y. Legallais** (*by invitation*), *National Cancer Institute, National Institute of Health, United States Public Health Service, Bethesda, Md.*
   The transparent chamber technique in the mouse in the study of tumor histophysiology.

FEDERATION "MIXER"

Wednesday, March 17, 9:00 p.m.

ARENA, CONVENTION HALL

Refreshments——Dancing

Admission by Registration Badge

THE AMERICAN PHYSIOLOGICAL SOCIETY

FIFTY-SEVENTH ANNUAL MEETING

PHYSIOLOGY A

Tuesday, March 16, 9 a.m.

BALLROOM, CONVENTION HALL

Renal Hemodynamics

1. **Francois C. Reubi** (*by invitation*), **Henry A. Schroeder, and Arnold H. Williams** (*by invitation*), *Department of Internal Medicine and the Oscar Johnson Institute, Washington University School of Medicine, and Barnes Hospital, St. Louis*
   Are there arterio-venous shunts in the kidney?
2. **J. H. Cort** (*by invitation*) **and Donald H. Barron,** *Laboratory of Physiology, Yale University School of Medicine*
   Neural control of the renal shunt.
3. **Domingo M. Gomez** (*introduced by* H. W. Smith), *New York University College of Medicine*
   Calculation of effective afferent and efferent renal resistance.
4. **Alfred A. Bolomey, Ernest F. Breed, Alexander Michie, Katherine Michie and Henry D. Lawson** (*introduced by* H. W. Smith), *New York University College of Medicine*
   Renal fraction in normal subjects and in subjects with essential hypertension.
5. **Woodrow Batten** (*by invitation*), **Ben C. Ogle** (*by invitation*), **Carlos Rapela** (*by invitation*) **J. Roy Hege, Jr., J. Maxwell Little and Harold D. Green,** *Department of Physiology and Pharmacology, Bowman Gray School of Medicine of Wake Forest College, Winston-Salem*
   Relationship between arterial pressure and renal blood flow.
6. **P. J. Talso** (*by invitation*), **A. P. Crosley, Jr.** (*by invitation*) **and R. W. Clarke,** *Medical Department Field Research Laboratory, Fort Knox*
   Effects of the cold pressor test on glomerular filtration and effective renal plasma flow.
7. **S. Baez** (*by invitation*), **A. Mazur and Ephraim Shorr,** *Department of Medicine, Cornell University Medical College and New York Hospital*
   Hepato-renal factors in circulatory homeostasis. XX. Antidiuretic action of liver VDM concentrates.
8. **H. L. White and Doris Rolf** (*by invitation*) *Department of Physiology, Washington University School of Medicine, St. Louis*
   Effects of exercise on renal circulation.
9. **J. P. Kriss, P. H. Futcher and M. L. Goldman** (*introduced by* H. A. Schroeder) *Department of Internal Medicine, Washington University School of Medicine, St. Louis*
   Influence of unilateral adrenalectomy and sympathectomy on homolateral renal function.
10. **David M. French, Pedro A. Molano and Walter M. Booker,** *Department of Pharmacology, Howard University School of Medicine, Washington*
    Renal function related to increased intra-abdominal pressure in anesthetized and unanesthetized dogs.
11. **Kendrick Hare, Luise Ungewitter** (*by invitation*), **Henry L. Barnett** (*by invitation*), **and Helen McNamara** (*by invitation*), *New York Hospital and the Departments of Pediatrics and Anatomy, Cornell University Medical College*
    Observations on the structure and function of the glomerulus in premature and full-term infants.
12. **W. G. Kubicek, R. B. Harvey and F. J. Kottke,** *University of Minnesota*
    The adrenalin sensitivity of the denervated dog kidney.

PHYSIOLOGY B

Tuesday, March 16, 9:00 a.m.

ROOM 20, CONVENTION HALL

Spinal Cord

1. **L. D. Carlson and A. W. Martin** (*with design in electronics by* R. S. Bark), *Dept. of Physiology and Biophysics, School of Medicine, University of Washington*
   A method for studying reflex activity under varying conditions.
2. **Jeane Siskel Frey** (*by invitation*) **and Robert Gesell,** *Physiology Laboratory, University of Michigan*
   A comparative study of the effects of several anticholinesterases on central nervous integration.
3. **A. van Harreveld and George A. Feigen** (*by invitation*), *California Institute of Technology, Pasadena*

The effect of nicotine on spinal synaptic conduction.

4. **David P. C. Lloyd and A. K. McIntyre** (*by invitation*), *Laboratories of The Rockefeller Institute for Medical Research, New York*
Potentials of dorsal roots and related phenomena.
5. **A. K. McIntyre** (*by invitation*) **and David P. C. Lloyd,** *Laboratories of The Rockefeller Institute for Medical Research, New York*
Spinal projection of hind-limb afferent fibers.
6. **Chandler McC. Brooks, J. C. Eccles** (*by invitation*) **and J. L. Malcolm** (*by invitation*), *Depts. of Physiology, University of Otago, Dunedin, N. Z., and the Johns Hopkins University*
Responses of inhibited motoneurones.
7. **J. E. Markee and Maude Williams** (*by invitation*), *Dept. of Anatomy, Duke University School of Medicine and Division of Physiology, Women's College of the University of North Carolina, Greensboro*
The separate contraction of functional areas in limb muscles during reflex withdrawal and crossed extension.
8. **Edward H. Lambert, Luis O. Mederos** (*by invitation*) **and Mavis P. Kelsey** (*by invitation*), *Sections on Physiology and Medicine, Mayo Clinic and Mayo Foundation*
A study of the ankle jerk in myxedema.
9. **Jerome Y. Lettvin** (*introduced by* W. S. McCulloch), *Dept. of Psychiatry, University of Illinois*
The path of suppression in the spinal grey matter.
10. **Irvin M. Korr and Martin J. Goldstein** (*by invitation*) *Still Memorial Research Trust, Kirksville, Mo.*
Dermatomal autonomic activity in relation to segmental motor reflex threshold.
11. **R. D. Teasdall** (*by invitation*), **and G. W. Stavraky,** *Dept. of Physiology, Faculty of Medicine, University of Western Ontario, London, Canada*
Response of deafferented neurones of spinal cord to impulses reaching them from higher levels of central nervous system.

PHYSIOLOGY C

Tuesday, March 16, 9:00 a.m.

Room 21, Convention Hall

Heart Electrophysiology

1. **Jane Sands Robb,** *Department of Pharmacology, Syracuse University College of Medicine*
Inconsistencies between experimental and theoretical electrocardiography.
2. **W. Kaufman** (*by invitation*), **H. M. Chernoff** (*by invitation*) **and L. H. Nahum,** *Laboratory of Physiology, Yale University School of Medicine*
A method of determining the spread of excitation in the ventricles of the dog's heart.
3. **L. H. Nahum, H. M. Chernoff** (*by invitation*) **and W. Kaufman** (*by invitation*), *Laboratory of Physiology, Yale University School of Medicine*
The nature of unipolar limb leads in the dog.
4. **H. M. Chernoff** (*by invitation*), **W. Kaufman** (*by invitation*) **and L. H. Nahum,** *Laboratory of Physiology, Yale University School of Medicine*
Derivation of leads I and III in the dog from analysis of unipolar limb leads.
5. **L. F. Nims, B. Kartin** (*by invitation*), **H. M. Chernoff** (*by invitation*) **and L. H. Nahum,** *Laboratory of Physiology, Yale University School of Medicine*
Heart temperature and its relation to the T-wave.
6. **Grace E. Wertenberger and Roberta Hafkesbring,** *Department of Physiology, Woman's Medical College of Pennsylvania*
The effect of serum hypersensitivity reactions upon the electrocardiogram.
7. **Alfred Leimdorfer,** *Department of Psychiatry, University of Illinois*
The effect of intravenous sodium cyanide on the electrocardiogram.
8. **Bruno Kisch, Franz M. Groedel** (*by invitation*) **and Paul R. Borchardt** (*by invitation*), *Biological Laboratory, Fordham University*
Differences in exocardial and endocardial electrograms of rabbit, dog and calf.
9. **William G. Turman** (*by invitation*) **and Jane Sands Robb,** *Department of Pharmacology, Syracuse University College of Medicine*
Experimental A-V block, method and discussion.
10. **Joseph H. Gast and Harold L. Dobson** (*introduced by* Allen D. Keller), *Department of Biochemistry, Baylor University College of Medicine, Houston*
Comparison of the effect of some steroids on the hearts of rats with choline deficiency.

PHYSIOLOGY D

Tuesday, March 16, 9:00 a.m.

Room 22, Convention Hall

Industrial Physiology

1. **William V. Whitehorn,** *Department of Physiology, Ohio State University*

Circulatory responses to exposure to barometric pressure of 30 mm. Hg.

2. **C. R. Spealman, E. W. Bixby** (*by invitation*), **J. LaRue Wiley** (*by invitation*), **Michael Newton** (*by invitation*) **and H. C. Bazett,** *Department of Physiology, University of Pennsylvania.*
Performance in warm environments following hemorrhage, albumin infusion, bed rest and exposure to cold.
3. **Henry Longstreet Taylor, Austin Henschel, Josef Brozek and Ancel Keys,** *Laboratory of Physiological Hygiene, University of Minnesota*
The behavior of certain characteristics related to exhausting work during recovery from semi-starvation.
4. **John Haldi and Winfrey Wynn** (*by invitation*), *Department of Physiology, Emory University, Ga.*
Industrial efficiency as affected by various kinds of foods consumed during the mid-morning and mid-afternoon rest periods.
5. **Austin Henschel, Henry Longstreet Taylor and Ancel Keys,** *Laboratory of Physiological Hygiene, University of Minnesota*
The effect of caloric level of refeeding on the recovery of work capacity following semi-starvation.
6. **Carleton B. Chapman** (*by invitation*), **Austin Henschel, John Minckler and Ancel Keys,** *Laboratory of Physiological Hygiene, University of Minnesota*
Effect of exercise on renal plasma flow.
7. **Albert A. Pollack and Earl H. Wood,** *Section on Physiology, Mayo Clinic, Mayo Foundation*
Venous pressure in the human leg during exercise and in various positions.
8. **A. T. Miller, Jr.,** *Laboratory of Applied Physiology, University of North Carolina*
The influence of oxygen administration on the cardiovascular response to exercise.
9. **Peter V. Karpovich and Nathan Millman** (*by invitation*), *Department of Physiology, Springfield College, Springfield, Mass.*
Seismography applied to the study of track running.
10. **W. W. Tuttle, Marjorie Wilson** (*by invitation*) **and Kate Daum** (*by invitation*), *Departments of Physiology and Nutrition, State University of Iowa*
The effect of low thiamine intake on the reaction time of women.
11. **John E. Peters** (*by invitation*) **and W. Horsley Gantt,** *Pavlovian Laboratory of the Phipps Psychiatric Clinic, Johns Hopkins Hospital*
Effect of graded degrees of muscular tension on human heart rate.

PHYSIOLOGY E

Tuesday, March 16, 9:00 a.m.

Room 17, Convention Hall

Respiration

1. **D. Saris** (*by invitation*), **E. A. Reed** (*by invitation*), **J. C. Scott,** *Department of Physiology, Hahnemann Medical College, Philadelphia*
Observations on the thoracic wall respiratory reflex.
2. **M. G. Larrabee and Robert Hodes,** *Johnson Foundation, University of Pennsylvania*
Cyclic changes in the respiratory center.
3. **Herbert L. Borison** (*by invitation*), **George Clark and S. C. Wang,** *Department of Physiology, College of Physicians and Surgeons, Columbia University*
Localization of certain spasmodic respiratory responses in the medulla oblongata of the cat.
4. **P. O. Chatfield and S. Mead** (*introduced by* E. M. Landis), *Department of Physiology, Harvard Medical School*
The role of the vagi in the crossed phrenic phenomenon.
5. **Joseph N. Spencer** (*by invitation*), **William B. Draper, Thomas M. Parry** (*by invitation*) **and Richard W. Whitehead,** *Department of Physiology and Pharmacology, University of Colorado Medical Center*
Studies on diffusion respiration. V. The hemoglobin-oxygen pump.
6. **Hermann Rahn and Arthur B. Otis,** *Department of Physiology, University of Rochester School of Medicine and Dentistry*
Man's respiratory response during acclimatization to high altitude.
7. **J. E. Geraci** (*by invitation*), **George E. Montgomery, Jr.** (*by invitation*) **and Earl H. Wood,** *Section on Physiology of the Mayo Clinic and Foundation*
Studies of arterial oxygen saturation in patients with suspected arterial hypoxemia, with use of a modified oximeter.
8. **Morton Galdston and Seymour A. Horwitz** (*introduced by* J. Murray Steele), *Department of Medicine, New York University College of Medicine and Research Service, Third (New York University) Medical Division, Goldwater Memorial Hospital*
Exchange of carbon dioxide and oxygen in the nasopharyngeal part of the respiratory dead space.
9. **Ward S. Fowler** (*introduced by* Julius H. Comroe, Jr.), *Department of Physiology and Pharmacology, Graduate School of Medicine,*

University of Pennsylvania
Respiratory dead space.

10. **F. N. Craig, Jane Stubbs** (*by invitation*) **and F. N. Marzulli** (*by invitation*), *Physiology Section, Medical Division, Army Chemical Center, Md.*
Analysis of combined effects of exercise and carbon dioxide inhalation on pulmonary ventilation in man.
11. **Donald H. Barron,** *Laboratory of Physiology, Yale University School of Medicine*
CO_2 content of maternal and fetal sheep blood.

PHYSIOLOGY F

Tuesday, March 16, 9:00 a.m.

Room 18, Convention Hall

Reproduction

1. **Arthur A. Cox,** (*introduced by* R. R. Overman), *Department of Physiology, University of Tennessee College of Medicine*
Observations on the oxytocic property of human blood.
2. **Annette LaBelle** (*by invitation*) **and Richard Tislow,** *Biological Research Laboratories, Schering Corporation, Bloomfield, N. J.*
Effect of lactogenic hormone on estrogen response of immature rats.
3. **Hubert R. Catchpole,** *Department of Pathology, University of Illinois College of Medicine*
Cell fractionation and gonadotrophin assays of anterior pituitary glands.
4. **M. X. Zarrow** (*by invitation*) **and W. T. Salter,** *Laboratories of Pharmacology and Toxicology, Yale University School of Medicine*
Comparison of biological and chemical evaluations of sex hormone balance. (Pharmacol.)
5. **F. D. Humm** (*by invitation*) **and W. T. Salter,** *Laboratories of Pharmacology and Toxicology, Yale University School of Medicine*
Chemical studies of sex steroid balance in human subjects.
6. **George Clark,** *Department of Anatomy, Chicago Medical School, and Yerkes Laboratories of Primate Biology, Orange Park, Fla.*
The lack of effect of estrogen on the sex skin of the infant male chimpanzee.
7. **José Gongora** (*by invitation*) **and Charles D. Kochakian,** *Department of Physiology and Vital Economics, University of Rochester*
The *in vitro* metabolism of testosterone to Δ^4-androstenedione-3,17 and cis-testosterone by rabbit liver homogenate.
8. **James H. Leathem,** *Bureau of Biological Research, Rutgers University*
High protein diets and testosterone propionate as related to plasma and liver proteins in rats.
9. **Boris B. Rubenstein,** *Department of Metabolism Endocrinology Research, Michael Reese Hospital, Chicago*
Vitamin E diminishes the vasomotor symptoms of menopause.
10. **S. R. M. Reynolds, L. M. Hellman** (*by invitation*) **and P. Bruns** (*by invitation*), *Carnegie Institution of Washington, Department of Embryology; and Department of Obstetrics, Johns Hopkins University School of Medicine*
Uterine contractions effective in dilating the human cervix, recorded by the multichannel strain gage tokodynamometer.
11. **David I. Macht,** *Division of Pharmacology, Sinai Hospital, Baltimore*
Concerning the nature of menstrual poison.
12. **Landrum B. Shettles and Zacharias Dische** (*by invitation*), *Depts. of Obstetrics and Gynecology, Anatomy and Biochemistry, College of Physicians and Surgeons, Columbia University*
The composition of polysaccharides of human cervical mucus.
13. **W. A. Selle and Otis B. Miller** (*by invitation*), *Depts. of Physiology and Dermatology, University of Texas Medical School, Galveston*
Fetal gastric secretion of radioiodine applied precutaneously to pregnant animals.

JOINT SESSION OF THE FEDERATION

Tuesday, March 16, 1:30 p.m.

Ballroom, Convention Hall

Program on page 314

PHYSIOLOGY BUSINESS MEETING

Tuesday, March 16, 4:15 p.m.

Room 20, Convention Hall

PHYSIOLOGY A

Wednesday, March 17, 9:00 a.m.

Ballroom, Convention Hall

Axone and Synapse

1. **Domingo Ampil** (*by invitation*) **and Harry Grundfest,** *Dept. of Neurology, College of Physicians and Surgeons, Columbia University*
Relation between synaptic delays and characteristics of preganglionic volleys initiating responses in cat autonomic ganglia.

2. **Harry Grundfest and Domingo Ampil** (*by invitation*), *Dept. of Neurology, College of Physicians and Surgeons, Columbia University*
   Preganglionic B fibers to the inferior mesenteric ganglion of the cat.
3. **Abraham M. Shanes,** *Dept. of Physiology and Biophysics, Georgetown University School of Medicine, Washington, Marine Biological Laboratory, Woods Hole, Mass., and Bermuda Biological Station*
   Medium concentration in relation to the water content and electrical properties of nerves.
4. **Gordon M. Schoepfle,** *Dept. of Physiology, Washington University School of Medicine, St. Louis*
   Nerve excitation as a function of membrane voltage.
5. **D. W. Bronk, F. Brink and M. G. Larrabee,** *Johnson Foundation, University of Pennsylvania*
   The sequence of functional changes in a neurone during narcosis and anoxia.
6. **J. M. Posternak** (*by invitation*) **and M. G. Larrabee,** *Johnson Foundation, University of Pennsylvania*
   Action of narcotics on synapses compared to action on axons in sympathetic ganglia.
7. **Robert Hodes,** *Dept. of Physical Medicine, Graduate School of Medicine, and the Eldridge Reeves Johnson Foundation, University of Pennsylvania*
   Conduction velocity of skeleto-motor nerve fibers to partially paralyzed muscles in human poliomyelitis.
8. **H. K. Hartline,** *Johnson Research Foundation, University of Pennsylvania*
   Retinal action potentials of photoreceptor cells and the discharge of nerve impulses in their axones.
9. **Francis O. Schmitt and Eduardo De Robertis** (*by invitation*), *Biology Department, Massachusetts Institute of Technology*
   Electron microscope observations of nerve structure.
10. **Rita Guttman,** *Brooklyn College*
    Resistance characteristics of rectifier element in single nerve fibers.

PHYSIOLOGY B

Wednesday, March 17, 9:00 a.m.

ROOM 20, CONVENTION HALL

Joint Session with The American Society for Experimental Pathology

See Pathology Program for titles

PHYSIOLOGY C

Wednesday, March 17, 9:00 a.m.

ROOM 21, CONVENTION HALL

Temperature Regulation

1. **Leon Eisenberg** (*by invitation*) **and H. C. Bazett,** *Physiology Department, University of Pennsylvania*
   Further observations on intravascular temperature in man.
2. **N. Kleitman, A. Ramsaroop** (*by invitation*), **and T. Engelmann** (*byi nvitation*), *Dept. of Physiology, University of Chicago*
   Variations in skin temperatures of the feet and hands and the onset of sleep.
3. **Michael Newton** (*introduced by* H. C. Bazett), *Dept. of Physiology, Medical School, University of Pennsylvania*
   Electrocardiographic changes on tilting.
4. **E. S. Fetcher, S. I. Rapaport** (*by invitation*) **and John F. Hall** (*by invitation*), *Aero Medical Laboratory, Wright Field*
   The physiological basis for the internal ventilation of clothing.
5. **S. I. Rapaport** (*by invitation*), **E. S. Fetcher and John F. Hall** (*by invitation*), *The Aero Medical Labovatory, Wright Field*
   Physiological protection of the extremities from severe cold.
6. **L. Love** (*by invitation*) **and H. C. Bazett,** *Dept. of Physiology, University of Pennsylvania*
   Heat loss and blood flow in the feet.
7. **Adelbert Ames, III** (*by invitation*), **Richard S. Griffith** (*by invitation*), **David A. Goldthwait** (*by invitation*), **Martin B. Macht** (*by invitation*), **and H. S. Belding,** *The Quartermaster Corps, Climatic Research Laboratory, Lawrence, Mass.*
   A study of various methods of rewarming men after exposure to extreme cold.
8. **E. F. Adolph,** *Dept. of Physiology, University of Rochester*
   Lethality of cold immersion in rats.
9. **Mortimer E. Bader, Martin B. Macht and Elizabeth L. Pillion** (*introduced by* H. S. Belding), *The Quartermaster Corps, Climatic Research Laboratory, Lawrence, Mass.*
   Peripheral vascular effects produced by localized warming of various skin areas.
10. **Martin B. Macht, Mortimer E. Bader and Elizabeth L. Pillion** (*introduced by* H. S. Belding), *The Quartermaster Corps, Climatic Research Laboratory, Lawrence, Mass.*

The effect of acetyl-beta-methyl-choline on blood flow through the hand at low temperatures.

11. **John R. Brobeck,** *Laby. of Physiology, Yale Univ. School of Medicine*
Food intake as a mechanism of temperature regulation in rats.

PHYSIOLOGY D

Wednesday, March 17, 9:00 a.m.

ROOM 22, CONVENTION HALL

Respiration

1. **R. W. Whitehead, D. L. G. Beshore** (*by invitation*), **W. B. Draper, Joseph N. Spencer** (*by invitation*) **and Thomas M. Perry** (*by invitation*), *Dept. of Physiology and Pharmacology, University of Colorado Medical Center*
Studies on diffusion respiration. VI. Changes in kidney function in dogs.
2. **Marshall Brucer** (*by invitation*) **and H. G. Swann,** *Dept. of Physiology, University of Texas Medical School*
A method of assaying the efficacy of resuscitating procedures.
3. **H. G. Swann and Marshall Brucer** (*by invitation*), *Dept. of Physiology, University of Texas Medical School*
The recovery period after resuscitation.
4. **A. Sokalchuk** (*by invitation*), **D. Ellis** (*by invitation*), **and E. M. Greisheimer,** *Dept. of Physiology, Temple University School of Medicine*
Pulmonary function as affected by operative positions.
5. **Nello Pace, Enrique Strajman** (*by invitation*), **and Elaine Walker** (*by invitation*), *Division of Medical Physics, University of California*
Influence of age on carbon monoxide desaturation in man.
6. **A. Hemingway, E. B. Brown, G. S. Campbell, F. Gollan and J. O. Elam,** *Dept. of Physiology, University of Minnesota*
Chronic hyperventilation in normal human subjects.
7. **Arthur B. Otis, Mitzi Suskind** (*by invitation*) **and Hermann Rahn,** *Dept. of Physiology, University of Rochester School of Medicine and Dentistry*
Effect of pressure breathing and posture upon the respiratory gas exchange and heart rate.
8. **Ralph W. Stacy** (*by invitation*), **Jack A. Hunter** (*by invitation*) **and F. A. Hitchcock,** *The Laboratory of Aviation Physiology, Ohio State University*
A mass spectrometer for the rapid, continuous analysis of respiratory gases.
9. **A. C. Barger** (*by invitation*), **G. S. Richardson** (*by invitation*), **and E. M. Landis,** *Dept. of Physiology, Harvard Medical School*
A Geiger-Müller counter system for tracer studies of gas exchanges in man.
10. **E. H. Wood, J. E. Geraci** (*by invitation*) **and D. L. Groom** (*by invitation*), *Section on Physiology of the Mayo Clinic and Foundation*
Photoelectric determination of blood oxygen saturation in man.
11. **George E. Montgomergy, Jr.** (*by invitation*), **J. E. Geraci** (*by invitation*) **and Earl H. Wood,** *Section on Physiology of the Mayo Clinic and Foundation*
Calibration of the Millikan compensated oximeter as used among white and colored persons.

PHYSIOLOGY E

Wednesday, March 17, 9:00 a.m.

ROOM 17, CONVENTION HALL

Enzymology

1. **Maurice M. Rapport** (*by invitation*), **Arda Alden Green and Irvine H. Page,** *Research Division of the Cleveland Clinic Foundation*
Enzymatic inactivation of serum vasoconstrictor.
2. **J. Maxwell Little.** *Dept. of Physiology and Pharmacology, Bowman Gray School of Medicine of Wake Forest College, Winston-Salem*
Two fractions of specific cholinesterase present in normal mouse brain.
3. **Charles D. Kochakian and Constance E. Stettner** (*by invitation*), *Dept. of Physiology and Vital Economics, University of Rochester*
A comparison of the effect of testosterone propionate and "growth hormone" on the body and organ weights and composition and the arginase and phosphatases of the kidney and liver of the castrated male mouse.
4. **R. H. De Meio** (*by invitation*), **A. E. Rakoff** (*by invitation*), **A. Cantarow and K. E. Paschkis,** *Jefferson Medical College, Philadelphia*
Mechanism of inactivation of α-estradiol by rat liver "in vitro".
5. **A. M. Freedman** (*by invitation*), **Alice Willis** (*by invitation*) **and H. E. Himwich,** *Medical Division, Army Chemical Center, Md.*
Correlation between signs of toxicity and cholinesterase level of brain and blood during recovery from di-isopropyl fluoro-phosphate (DFP) poisoning.
6. **S. Spiegelman and John M. Reiner,** *Dept. of*

Bacteriology and Immunology, Washington University School of Medicine, St. Louis
Protection by substrate against inhibition of enzymatic adaptation by protein denaturants.

7. **Clary J. Fischer** (*by invitation*) **and Roy O. Greep,** *Harvard School of Dental Medicine*
Effect of magnesium on alkaline phosphatase as influenced by pH, enzyme concentration and aging.

8. **Emily A. Feld** (*by invitation*), **Mortimer A. Rothenberg** (*by invitation*) **and David Nachmansohn,** *Dept. of Neurology, College of Physicians and Surgeons, Columbia University.*
On the mechanism of the action of di-isopropyl fluorophosphate.

9. **John M. Reiner and S. Spiegelman,** *Dept. of Bacteriology and Immunology, Washington University School of Medicine, St. Louis*
The partial purification and some properties of an adaptation-stimulating principle from yeast.

10. **Otto H. Müller,** *Dept. of Physiology, Syracuse University College of Medicine*
A polarographic study of the action of carbonic anhydrase at the dropping mercury electrode.

PHYSIOLOGY F

Wednesday, March 17, 9:00 a.m.

ROOM 18, CONVENTION HALL

Hypertension and Kidney

1. **George E. Brown, Jr.** (*by invitation*) **and Earl H. Wood,** *Section on Physiology of the Mayo Clinic and Foundation*
Effect of tetra-ethyl-ammonium chloride on intra-arterial blood pressure in patients with coarctation of the aorta.

2. **Irvine H. Page, R. D. Taylor and J. J. Reinhard** (*by invitation*), *Research Division of the Cleveland Clinic Foundation*
The influence of tetraethyl ammonium, hepatectomy and selective destruction of the nervous system on vascular reactivity.

3. **E. A. Ohler** (*by invitation*) **and G. E. Wakerlin,** *Dept. of Physiology, University of Illinois College of Medicine*
Further studies on the treatment of experimental renal hypertension with paredrine hydrobromide.

4. **Arnold H. Williams** (*by invitation*) **and Henry A. Schroeder,** *Dept. of Internal Medicine and the Oscar Johnson Institute, Washington University School of Medicine, and Barnes Hospital, St. Louis*
The asystolic arterial pressure gradient as a measure of local peripheral resistance.

5. **Norman S. Olsen** (*by invitation*), **Henry A. Schroeder and Melvin L. Goldman** (*by invitation*), *Depts. of Biological Chemistry and Internal Medicine and the Oscar Johnson Institute, Washington University School of Medicine, and Barnes Hospital, St. Louis*
Methods for extraction of pressor substances from hypertensive blood.

6. **C. A. Schaffenburg** (*by invitation*) **and H. Selye,** *Institut de Medecine et de Chirurgie Experimentales, Universite de Montreal*
Blood electrolite changes in experimental renal hypertension.

7. **Edwin P. Hiatt,** *Dept. of Physiology, University of North Carolina School of Medicine*
Effects of oral cinchona alkaloids on circulation of dogs with experimental hypertension.

8. **Helene Wastl,** *Dept. of Anatomy, Hahnemann Medical College, Philadelphia*
Influence of tea from leaves of ceanothus americanus on blood pressure of hypertensive rats.

9. **B. W. Zweifach, S. Rosenfeld** (*by invitation*) **and Ephraim Shorr,** *Dept. of Medicine, Cornell University Medical College and the New York Hospital*
Hepato-renal factors in circulatory homeostasis. XVI. Vascular changes in mesentery in renal hypertension in rats.

10. **Eric Ogden,** *Dept. of Physiology, University of Texas Medical Branch*
Influence of dibenamine on renal function of the dog.

11. **Arthur J. Dziemian** (*introduced by* W. Fleischmann) *Medical Division, Army Medical Center, Md.*
The effects of burns on kidney function.

12. **J. T. Bradbury, O. F. Kraushaar** (*by invitation*), **W. E. Brown,** (*by invitation*), *Dept. of Obstetrics and Gynecology, University of Iowa*
The effect of morphine on urinary volume in women.

PHYSIOLOGY A

Wednesday, March 17, 1:45 p.m.

BALLROOM, CONVENTION HALL

Nerve Metabolism

1. **J. Magnes and A. Geiger** (*introduced by* H. Grundfest), *Dept. of Physiology, Hebrew University, Jerusalem, and Dept. of Psychiatry, College of Physicians and Surgeons, Columbia University*
Effect of hydrogen and bicarbonate ions on the metabolic rate of the perfused cat brain.

2. **Seymour S. Kety, Francis D. W. Lukens,**

Rachel B. Woodford (*by invitation*), **Merel H. Harmel** (*by invitation*), **F. A. Freyhan** (*by invitation*) **and Carl F. Schmidt,** *Depts. of Pharmacology, Medicine and Psychiatry, University of Pennsylvania and the Delaware State Hospital*

The effects of insulin hypoglycemia and coma on human cerebral metabolism and blood flow.

3. **F. D. Carlson** (*by invitation*), **F. Brink and D. W. Bronk,** *Johnson Foundation, University of Pennsylvania*

A method for direct measurement of rate of oxygen utilization by nerve.

4. **C. M. Connelly** (*by invitation*) **and D. W. Bronk,** *Johnson Foundation, University of Pennsylvania*

Measurements of rapid changes in oxygen consumption by nerve following brief periods of stimulation.

5. **R. G. Grenell, P. W. Davies** (*by invitation*), **and D. W. Bronk,** *Johnson Foundation, University of Pennsylvania*

The effects of brain anemia on cortical oxygen consumption in vivo.

6. **P. W. Davies** (*by invitation*), **R. G. Grenell and D. W. Bronk,** *Johnson Foundation, University of Pennsylvania*

The time course of in-vivo oxygen consumption of cerebral cortex following electrical stimulation.

7. **L. L. Boyarski, S. Postel and A. Rosenblatt** (*introduced by* R. W. Gerard), *Dept. of Physiology, University of Chicago*

Enzyme inhibitors on conduction and respiration of frog nerve.

8. **V. B. Brooks and R. E. Ransmeier** (*introduced by* R. W. Gerard), *Dept. of Physiology, University of Chicago.*

Enzyme inhibitors on electric activity of frog brain.

9. **C. Haber and L. Saidel** (*introduced by* R. W. Gerard), *Dept. of Physiology, University of Chicago*

Glutamic acid in neural activity.

10. **B. Libet,** *Dept. of Physiology, University of Chicago*

Adenosinetriphosphatase in nerve.

PHYSIOLOGY B

Wednesday, March 17, 1:45 p.m.

ROOM 20, CONVENTION HALL

Gastric Secretion

1. **T. L. Patterson, J. Kaulbersz, D. J. Sandweiss and H. C. Saltzstein** (*by invitation*), *Depts. of Physiology and Surgery, Wayne Univ. College of Medicine, and Harper Hospital, Detroit*

The effect of oöphorectomized dogs' urine extract on gastric secretion.

2. **C. R. Robertson** (*by invitation*) **and M. I. Grossman,** *Dept. of Clinical Science, Univ. of Illinois College of Medicine*

Potentiation of the gastric secretory response to histamine by parasympathomimetic drugs.

3. **A. Littman** (*by invitation*) **and M. I. Grossman,** *Dept. of Clinical Science, Univ. of Illinois College of Medicine*

Action of antacids in the human stomach; results with zirconium phosphate.

4. **M. I. Grossman, C. R. Robertson** (*by invitation*), **and W. L. Anderson** (*by invitation*). *Dept. of Clinical Science, Univ. of Illinois College of Medicine*

Inhibition of gastric secretion by "histaminase".

5. **E. Hale** (*by invitation*) **and M. I. Grossman,** *Dept. of Clinical Science, Univ. of Illinois College of Medicine*

Studies on the mechanism of "acid rebound" in gastric acidity.

6. **Franklin Hollander and Frances U. Lauber** (*by invitation*), *Gastroenterology Research Laboratory, Mt. Sinai Hospital, New York City*

The pH of gastric mucous secretion after equilibration in vitro with alveolar air.

7. **Gladys R. Bucher and Arthur Anderson** (*by invitation*), *Dept. of Biochemistry, Vanderbilt University*

The uropepsin output in cats given histamine-caffeine in beeswax.

8. **S. A. Komarov, Harry Shay and Herman Siplet** (*by invitation*), *Fels Research Institute, Temple University School of Medicine*

Secretion of mucin in response to sham feeding and histamine stimulation.

9. **Leslie E. Edwards and Carolyn Trowbridge Edwards** (*introduced by* R. W. Ramsey), *Dept. of Physiology and Pharmacology, Medical College of Virginia*

A method for the study of gastric secretion in vitro.

10. **Carolyn Trowbridge Edwards and Leslie E. Edwards** (*introduced by* R. W. Ramsey), *Dept. of Physiology and Pharmacology, Medical College of Virginia*

Factors influencing in-vitro secretion of pepsin.

11. **Charles A. Winter and Charles W. Mushett** (*by invitation*), *Merck Institute for Therapeutic Research, Rahway*

Gastrointestinal and hematological responses in dogs to large doses of histamine and an antihistaminic drug.

12. **A. H. Ryan,** *Dept. of Physiology and Pharmacology, Chicago Medical School*
Sympathetic reaction in peptic ulcer.
13. **J. Katz** (*by invitation*), **Robert L. Dryer** (*by invitation*), **W. D. Paul** (*by invitation*) **and J. I. Routh,** *Depts. of Biochemistry and Internal Medicine, College of Medicine, Univ. of Iowa*
Isolation and assay of an anti-secretory substance from duodenal mucosa. (Biochem.)

PHYSIOLOGY C

Wednesday, March 17, 1:45 p.m.

ROOM 21, CONVENTION HALL

Temperature Regulation

1. **V. E. Hall, R. Grant** (*by invitation*) **and J. Field,** *Dept. of Physiology, Stanford University*
The influence of substances affecting body temperature on thermal polypnea.
2. **J. Field, C. N. Peiss** (*by invitation*), **and V. E. Hall,** *Dept. of Physiology, Stanford University*
The influence of substances affecting body temperature on oxygen consumption and glycolysis in brain.
3. **Walter C. Randall, Douglas E. Smith and Alrick B. Hertzman,** *Dept. of Physiology, St. Louis University School of Medicine*
Some cutaneous responses to "reflex heating".
4. **Douglas E. Smith, Walter C. Randall and Alrick B. Hertzman,** *Dept. of Physiology, St. Louis University School of Medicine*
Some cutaneous responses to "reflex cooling'.
5. **Walter B. Shelley, Peter N. Horvath** (*by invitation*), **and Steven M. Horvath,** *Depts. of Dermatology and Physical Medicine, Univ. of Pennsylvania School of Medicine.*
Inhibition of sweating by means of iontophoresis.
6. **Steven M. Horvath,** *Dept. of Physical Medicine, Graduate School of Medicine, University of Pennsylvania*
Orthostatic hypotension following hot or cold baths.
7. **J. F. Herrick and J. L. Bollman,** *Div. of Experimental Medicine, Mayo Foundation*
The effect of hyperthermia on the flow of thoracic duct lymph.
8. **James L. A. Roth and John A. Frantz** (*introduced by* J. W. Heim), *Physiology Branch, Aero Medical Laboratory, Wright Field*
Metabolic balances in the cold environment: I. Nitrogen and water exchanges.
9. **John A. Frantz and James L. A. Roth** (*introduced by* J. W. Heim), *Physiology Branch, Aero Medical Laboratory, Wright Field*
Metabolic balances in the cold environment: II. Energy exchanges.
10. **S. Rodbard and F. Sampson** (*by invitation*), *Cardiovascular Dept., Research Institute, Michael Reese Hospital, Chicago*
Further evidence for a temperature-sensitive mechanism in a poikilotherm (the turtle).
11. **Harold D. Green and Ben C. Ogle** (*by invitation*), *Dept. of Physiology and Pharmacology, Bowman Gray School of Medicine of Wake Forest College*
Vasodilation produced by etamon, priscol, body warming and spinal anesthesia in normal extremities.

PHYSIOLOGY D

Wednesday, March 17, 1:45 p.m.

ROOM 22, CONVENTION HALL

Shock

1. **Samuel A. Corson and Elizabeth O'Leary, with the technical assistance of Opal Cain,** *Depts. of Physiology, School of Medicine, Univ. of Minnesota and Howard University*
The influence of sodium salts on the extracellular space in experimental hypoproteinemic edema.
2. **R. R. Overman, C. P. Tharp** (*by invitation*) **and A. H. Tuttle** (*by invitation*), *Dept. of Physiology, Univ. of Tennessee College of Medicine*
Alterations in fluid and ionic distribution in various patho-physiological conditions.
3. **C. P. Tharp** (*introduced by* R. R. Overman, *Dept. of Physiology, University of Tennessee College of Medicine*
Comparison of mannitol and thiocyanate volumes in several patho-physiological conditions.
4. **N. R. Joseph** (*by invitation*), **C. I. Reed and I. E. Stock** (*by invitation*), *Dept. of Physiology, College of Medicine, and Dept. of Chemistry, College of Pharmacy, University of Illinois*
Electrochemical determination of ionic diffusion through the synovial membrane.
5. **E. W. Bixby** (*introduced by* H. C. Bazett), *Physiology Dept., Medical School, University of Pennsylvania*
The recovery of sodium thiocyanate from whole blood and plasma as related to the measurement of extracellular fluid volumes.
6. **W. D. Collings, C. J. Martin** (*by invitation*) **and G. R. Walters** (*by invitation*), *Dept. of Physiology, State University of Iowa*

B & Z Library

A method for constant, long-term intravenous infusion of the unanesthetized dog.

7. **John S. Kirk** (*introduced by* K. E. Jochim), *Dept. of Physiology, Univ. of Kansas*
   Visceral capillary permeability changes produced in the dog by anaphylactic shock and venous statis.
8. **Parke H. Woodard** (*introduced by* K. E. Jochim), *Dept. of Physiology, Univ. of Kansas*
   Anaphylactic shock in a restricted circulation system excluding the liver in the dog.
9. **Louis Moreau** (*by invitation*), **Marvin Balistocky** (*by invitation*), **and L. V. Heilbrunn,** *Zoological Laboratory, University of Pennsylvania*
   Studies of shock in frogs: I. Shock due to electrical injury.
10. **Monica Reynolds** (*introduced by* Magnus I. Gregersen), *Dept. of Physiology, College of Physicians and Surgeons, Columbia University*
    An analysis of the effects of large volumes of isotonic saline on the circulation of severely hemorrhaged dogs.
11. **R. C. Ingraham and Franklin Roemhild** (*by invitation*), **and H. Goldberg** (*by invitation*) *Dept. of Physiology, Univ. of Illinois College of Medicine*
    Further observations on the effect of pentobarbital and of an adrenolytic agent upon the survival of animals subjected to a procedure resulting in experimental hemorrhagic shock.
12. **J. Garrott Allen and Willadene Egner** (*by invitation*), *Dept. of Surgery, University of Chicago*
    The reaction caused by the intravenous injection of salmine sulfate in dogs.

PHYSIOLOGY E

Wednesday, March 17, 1:45 p.m.

Room 17, Convention Hall

Carbohydrate Metabolism

1. **J. Clifford Stickney, David W. Northup and Edward J. Van Liere,** *Dept. of Physiology, West Virginia University School of Medicine*
   Effect of anoxia on the glucose tolerance curve of dogs.
2. **Edward J. Van Liere, J. Clifford Stickney and David W. Northup,** *Dept. of Physiology, West Virginia University School of Medicine*
   Effect of acclimatization on blood sugar response to anoxia.
3. **M. Wierzuchowski** (*introduced by* William H. Chambers), *Physiological Institute of the Medical Faculty, University of Lodz, Poland*
   The effect of body temperature on the respiratory transformation of intravenous carbohydrates in amytal-anesthetized dogs.
4. **Roger M. Reinecke and Paul J. Hauser** (*by invitation*), *University of Minnesota*
   Arterio-venous blood sugar studies on the kidney of the eviscerated dog.
5. **C. H. Beatty** (*introduced by* Magnus I. Gregersen), *Dept. of Physiology, College of Physicians and Surgeons, Columbia University*
   The femoral A-V glucose differences following glucose infusion in unanesthetized normal and adrenalectomized dogs.
6. **R. C. de Bodo, I. H. Slater** (*by invitation*), **H. F. Weisberg** (*by invitation*), **K. F. Prescott** (*by invitation*), *Dept. of Pharmacology, New York University College of Medicine*
   Adrenaline hyperglycemia in hypophysectomized dogs.
7. **M. Perlmutter** (*introduced by* G. W. Thorn) **and R. O. Greep,** *Dept. of Medicine, Harvard Medical School; the Medical Clinic, Peter Bent Brigham Hospital; and the Harvard School of Dental Medicine*
   Effect of insulin upon the in vitro glucose utilization and glycogenesis of the diaphragm of normal and pituitarectomized rats. Adaptation of this technique as an assay for serum content of insulin and anti-insulin substances.
8. **Knud Lunbaek and James Stevenson** (*introduced by* C. N. H. Long). *Dept. of Physiological Chemistry and Laby. of Physiology, Yale University School of Medicine.*
   The effect of previous carbohydrate deprivation on the carbohydrate metabolism of isolated muscle.
9. **Lawrence Greenman and John C. Rathbun** (*introduced by* T. S. Danowski), *Dept. of Research Medicine, University of Pittsburgh School of Medicine, the Renziehausen Foundation and the Children's Hospital of Pittsburgh*
   Studies of galactose and glucose metabolism.
10. **J. D. Meyers** (*introduced by* E. A. Stead, Jr.), *Dept. of Medicine, Duke University School of Medicine*
    The effects of the intravenous administration of glucose and amino acids on the hepatic blood flow and splanchnic oxygen consumption of man.
11. **T. S. Danowski,** *Dept. of Research Medicine and the Renziehausen Foundation, University of Pittsburgh School of Medicine*
    Alterations in the rate of glycolysis in human blood following the addition of salts.

PHYSIOLOGY F

Wednesday, March 17, 1:45 p.m.

ROOM 18, CONVENTION HALL

Endocrinology

1. **R. de Grandpre** (*by invitation*), **J. L. Prado** (*by invitation*), **P. Dontigny** (*by invitation*), **J. Leduc** (*by invitation*) **and H. Selye,** *Institut de Medecine et de Chirurgie Experimentales, Universite de Montreal, Canada*
   Influence of protein hydrolysates on the production of nephrosclerosis and hypertension by anterior-pituitary preparations.
2. **Henry D. Lauson, Howard A. Eder** (*by invitation*), **Francis P. Chinard** (*by invitation*), **George C. Cotzias** (*by invitation*) **and Roger L. Greif** (*by invitation*), *Hospital of the Rockefeller Institute for Medical Research*
   Estimation of the rate of antidiuretic hormone secretion in normal man.
3. **William O. Maddock, Edwin S. Jungck** (*by invitation*), **and Carl G. Heller,** *Dept. of Physiology, University of Oregon Medical School*
   Antigonadotrophin formation to sheep FSH: effectiveness against endogenous gonadotrophic hormones in men.
4. **Savino A. D'Angelo, Albert S. Gordon and Harry A. Charipper,** *Dept. of Biology, Washington Square College of Arts and Sciences, New York University*
   The effect of inanition on pituitary-adrenal function in the guinea pig.
5. **David Rapport, Attilio Canzanelli and Ruth Guild** (*by invitation*), *Dept. of Physiology, Tufts College Medical School*
   Effects of the ablation of the pituitary, adrenals and thyroid on liver regeneration, nucleic acid partition, etc.
6. **I. Arthur Mirsky, John Wachman** (*by invitation*), **C. J. Podore** (*by invitation*) **and R. H. Broh-Kahn** (*by invitation*), *May Institute for Medical Research of the Jewish Hospital, Cincinnati*
   Insulin excretion in normal man.
7. **R. H. Broh-Kahn** (*by invitation*) **and I. Arthur Mirsky,** *May Institute for Medical Research of the Jewish Hospital, Cincinnati*
   Insulin inactivation by tissue extracts.
8. **I. H. Slater** (*by invitation*), **R. C. de Bodo, H. F. Weisberg** (*by invitation*) **and S. P. Kiang** (*by invitation*), *Dept. of Pharmacology, New York University College of Medicine*
   A study of insulin hypersensitivity in hypophysectomized dogs.
9. **William T. Salter, MacAllister W. Johnston** (*by invitation*) **and Janet Beach** (*by invitation*), *Laboratories of Pharmacology and Toxicology, Yale Univ. School of Medicine*
   Extra-thyroidal iodine.
10. **Warren O. Nelson and Helen E. Wheeler** (*by invitation*), *Dept. of Anatomy, University of Iowa College of Medicine*
    Effect of iodine in hypothyroid rats.
11. **S. B. Barker and H. J. Lipner** (*by invitation*), *Dept. of Physiology, State University of Iowa*
    Determination of protein-bound iodine in biological material.
12. **Willie W. Smith,** *Laboratory of Physical Biology, National Institute of Health, Bethesda, Md.*
    The influence of thiourea and thiouracil on the response of rats to methyl chloride.
13. **Albert S. Gordon and Grace F. Katsh** (*by invitation*), *Dept. of Biology, Washington Square College of Arts and Sciences, New York University*
    The relation of the endocrine gland system to macrophagic activity.

PHYSIOLOGY EDITORIAL CONFERENCE

Wednesday, March 17, 7:30 p.m.

ROOM 17, CONVENTION HALL

A. C. Ivy, *Chairman*

All members interested in the publication affairs of the Society are cordially invited to attend.

Federation "Mixer"

Wednesday, March 17, 9:00 p.m.

ARENA, CONVENTION HALL

PHYSIOLOGY A

Thursday, March 18, 9:00 a.m.

BALLROOM, CONVENTION HALL

Hypertension

1. **Isabelle Dougherty and Sister M. A. C. Day** (*introduced by* A. B. Hertzman), *Dept. of Physiology, St. Louis University School of Medicine*
   Observations on the peripheral circulation in neurogenic hypertension.
2. **W. G. Moss** (*by invitation*) **and G. E. Wakerlin,** *Dept. of Physiology, University of Illinois College of Medicine*
   The role of the nervous system in experimental hypertension in the dog.
3. **G. E. Wakerlin, John Marshall, Hiroski Minatoya, Rufus Walker and A. Kaplan,** *Dept. of Physiology, University of Illinois College of Medicine*

Further studies on the treatment and prophylaxis of experimental renal hypertension with renal extracts.

4. **Robert D. Taylor, A. C. Corcoran and Irvine H. Page**, *Research Division of the Cleveland Clinic Foundation*
   Effects of denervation on experimental renal hypertension.
5. **Lena A. Lewis and Irvine H. Page,** *Research Division of the Cleveland Clinic Foundation*
   Nature of increased serum β-globulin content in malignant hypertension and diabetic retinopathy.
6. **Ronald E. Scantlebury** (*introduced by* T. L. Patterson), *Dept. of Physiology and Pharmacology, University of Arkansas School of Medicine*
   Development of chronic hypertension in dogs by the occlusion of the carotid arteries.
7. **Melvin L. Goldman** (*by invitation*) **and Henry A. Schroeder,** *Dept. of Internal Medicine and the Oscar Johnson Institute, Washington University School of Medicine, and Barnes Hospital, St. Louis*
   The immediate pressor effect of desoxycorticosterone acetate in hypertensive and normotensive subjects.
8. **Robert E. Shipley and O. M. Helmer,** *Lilly Laboratory for Clinical Research, Indianapolis General Hospital*
   Observations on the sustained pressor principle in different animal species.
9. **O. M. Helmer, Robert E. Shipley and K. G. Kohlstaedt,** *Lilly Laboratory for Clinical Research, Indianapolis General Hospital*
   Further studies on the identity of the sustained pressor principle.
10. **Henry A. Schroeder, Melvin L. Goldman** (*by invitation*) **and Norman S. Olsen** (*by invitation*), *Depts. of Internal Medicine and Biological Chemistry and the Oscar Johnson Institute, Washington University School of Medicine, and Barnes Hospital, St. Louis*
    Pressor substances in hypertensive blood.
11. **Raymond E. Weston, Leon Hellman** (*by invitation*), **Doris J. W. Escher** (*by invitation*) **and Louis Leiter** (*by invitation*), *Medical Division, Montefiore Hospital, New York*
    Effect of low sodium and Kempner diets on renal hemodynamics and electrolyte excretion in hypertensives.
12. **L. Horlick** (*by invitation*) **and L. N. Katz** *Cardiovascular Dept., Research Institute, Michael Reese Hospital, Chicago*
    Standardization of a method for the production of experimental atherosclerosis in the chicken.

PHYSIOLOGY B

Thursday, March 18, 9:00 a.m.

Room 20, Convention Hall

Axone and Synapse Action

1. **Otto H. Schmitt,** *Depts. of Zoology and Physics, University of Minnesota*
   Measurement of electrical energy release "impedance" and longitudinal transport in nerve by differential electrode techniques.
2. **R. Beutner and T. C. Barnes,** *Dept. of Pharmacology, Hahnemann Medical College and Hospital of Philadelphia*
   A new concept of phase-boundary potential applied to the electrophysiology of nerve.
3. **Birdsey Renshaw and Herbert Rosenbaum** (*by invitation*), *Dept. of Physiology, University of Oregon Medical School*
   Does injury to an axon promptly induce altered excitability in its cell of origin?
4. **Mortimer A. Rothenberg** (*introduced by* David Nachmansohn), *Dept. of Neurology, College of Physicians and Surgeons, Columbia University*
   Rate of penetration of electrolytes into nerve fibers.
5. **Wynne Sharples** (*by invitation*), **Harry Grundfest and David Nachmansohn,** *Dept. of Neurology, College of Physicians and Surgeons, Columbia University*
   Adenosinetriphosphatase and conduction of the nerve impulse.
6. **David Nachmansohn and Mortimer A. Rothenberg** (*by invitation*), *Dept. of Neurology, College of Physicians and Surgeons, Columbia University*
   The difference between pharmacological action and the physiological role of acetylcholine
7. **James E. P. Toman,** *Dept. of Physiology, University of Utah School of Medicine*
   Some effects of anticholinesterases upon frog sciatic nerve.
8. **Leonard H. Elwell** (*by invitation*) **and John W. Bean,** *Physiology Laboratory, University of Michigan*
   Effects of positive intrapulmonic pressure on muscular contraction.
9. **W. E. Burge,** *Dept. of Physiology, University of Illinois*
   Consciousness and unconsciousness during anesthesia in relation to brain potential.

PHYSIOLOGY C

Thursday, March 18, 9:00 a.m.

Room 21, Convention Hall

Aviation Physiology

1. **E. L. Corey,** *Physiological Laboratory, University of Virginia Medical School*
   Pilot metabolism and respiratory activity during varied flight tasks.
2. **M. Mason Guest,** *Wayne University College of Medicine*
   Diet and gastro-intestinal symptoms at altitude.
3. **S. W. Britton and V. A. Pertzoff** (*by invitation*), *Physiological Laboratory, University of Virginia*
   Circulatory reserves shown by animals under acceleratory exposure.
4. **Milton Joffe** (*by invitation*) **and F. A. Hitchcock,** *Laboratory of Aviation Physiology, Ohio State University*
   Studies on deceleration.
5. **H. Jacobs** (*by invitation*), **A. Karstens** (*by invitation*), **and J. P. Henry,** *Aero Medical Laboratory, Air Materiel Command, Wright Field*
   The effect of pressure breathing on circulating blood volume. (2) Distension of the venous arteries.
6. **J. L. Gamble** (*by invitation*), **R. S. Shaw** (*by invitation*), **O. Gauer** (*by invitation*) **and J. P. Henry,** *Aero Medical Laboratory, Air Materiel Command, Wright Field*
   Studies of the pathological physiology of negative "G" in animals and man.
7. **R. S. Shaw** (*by invitation*), **J. L. Gamble, Jr.** (*by invitation*), **P. J. Maher** (*by invitation*), **J. P. Henry and O. Gauer** (*by invitation*), *Aero Medical Laboratory, Air Materiel Command, Wright Field*
   On the use of venous pressure in the head as a tolerance index of negative "G" in humans.
8. **Samuel Gelfan and George D. Davis** (*by invitation*), *Laboratory of Physiology, Yale University School of Medicine*
   Explosive decompression of monkeys at extreme altitides.
9. **Madeline Fusco** (*by invitation*), **Henry Mellette** (*by invitation*) **and Fred A. Hitchcock,** *Laboratory of Aviation Physiology, Ohio State University*
   Determination of the etiology of pathological effects of explosive decompression on the lungs of rats.
10. **Fred A. Hitchcock, Floyd M. Beman** (*by invitation*) **and John A. Kemph** (*by invitation*), *Laboratory of Aviation Physiology, Ohio State University*
    Estimation of subcutaneous pressure in animals explosively decompressed to pressures of 30 mm. Hg.
11. **True W. Robinson** (*introduced by* J. W. Heim), *Physiology Branch, Aero Medical Laboratory, Wright Field*
    Pretreatment against decompression sickness.

PHYSIOLOGY D

Thursday, March 18, 9:00 a.m.

ROOM 22, CONVENTION HALL

General Physiology

1. **F. Brink and J. M. Posternak** (*by invitation*), *Johnson Foundation, University of Pennsylvania*
   Thermodynamic analysis of the relative effectiveness of narcotics.
2. **Mary Cregar** (*introduced by* L. V. Heilbrunn), *Zoological Laboratory, University of Pennsylvania*
   The effect of anesthetics on calcium release.
3. **A. L. Hopkins** (*introduced by* L. V. Heilbrunn), *Marine Biological Laboratory, Woods Hole*
   Autotomy as a test for toxic factor.
4. **J. Painter and C. M. Pomerat,** *Tissue Culture Laboratory of the Department of Anatomy and the Psychopathetic Hospital, Medical Branch, University of Texas*
   The toxicity of various stimulants of nervous tissue as revealed by tissue culture technique.
5. **L. E. Chadwick and V. G. Dethier** (*by invitation*), *Medical Division, Army Chemical Corps, Army Chemical Center, Md., and Johns Hopkins University*
   The distribution of response thresholds in studies of insect chemoreception.
6. **Wilhelm Buschke,** *Ayer Foundation Ophthalmic Research Laboratory, Manhattan Eye, Ear and Throat Hospital*
   Epithelial movements in woundhealing in frog corneas.
7. **Alfred F. Bliss,** *Dept. of Physiology, Tufts College Medical School*
   A light-stable visual purple
8. **J. H. Bodine and L. R. Fitzgerald** (*by invitation*), *Zoological Laboratory, State University of Iowa*
   Change in riboflavin during embryonic development.
9. **J. Percy Baumberger, George F. Leong** (*by invitation*) **and Kathleen Bardwell** (*by invitation*), *Physiology Dept., School of Medicine, Stanford University*
   A further study of the effect of hemoglobin on cellular respiration.
10. **C. C. Hassett** (*introduced by* L. E. Chadwick), *Medical Division, Army Chemical Corps, Army Chemical Center, Md.*
    The utilization of carbohydrates by drosophila melanogaster.
11. **John O. Hutchins, Betty Podolsky** (*by invitation*) **and T. M. McMahon** (*by invitation*),

Toxicity Laboratory and Dept. of Physiology, University of Chicago

Effects of tris-(β-chloroethyl) amine on respiration, carbohydrate and protein synthesis, and cell division in *Chilomonas*.

12. **V. G. Dethier** (*by invitation*) **and L. E. Chadwick,** *Johns Hopkins University and the Medical Division, Army Chemical Corps, Army Chemical Center, Md.*

The stimulating effect of glycols and their polymers on the tarsal receptors of blowflies.

PHYSIOLOGY E

Thursday, March 18, 9:00 a.m.

Room 17, Convention Hall

Metabolism

1. **Philip Dow and Robert H. Shuler** (*by invitation*), *Dept. of Physiology, University of Georgia School of Medicine*

The basal metabolic rate of Georgia medical students.

2. **Lawrence R. Prouty** (*by invitation*), **Martha J. Barrett** (*by invitation*) **and James D. Hardy,** *New York Hospital and the Dept. of Physiology, Cornell University Medical College*

A simple calorimeter for simultaneous determination of heat production and heat loss in laboratory animals.

3. **Grace M. Roth and Charles Sheard,** *Section on Physiology and Division of Physics and Biophysical Research, Mayo Clinic and Mayo Foundation*

Relation of basal metabolic rate to vasodilatation and vasoconstriction of extremities of normal subjects as measured by skin temperatures.

4. **Jack L. Strominger** (*by invitation*) **and John R. Brobeck,** *Laboratory of Physiology, Yale University School of Medicine*

Adjustment of caloric intake of rats to dietary change.

5. **A. Lane** (*by invitation*) **and A. C. Ivy,** *Dept. of Clinical Science, University of Illinois College of Medicine*

Studies on the nutritional effects of heated fats.

6. **M. E. Hanson** (*by invitation*) **and M. I. Grossman,** *Dept. of Clinical Science, University of Illinois College of Medicine*

The failure of intravenous glucose to inhibit food intake in dogs.

7. **H. C. Meng** (*by invitation*) **and Smith Freeman,** *Dept. of Physiology and Dept. of Experimental Medicine, School of Medicine, Northwestern University*

Preliminary study of complete parenteral alimentation.

8. **Hazel E. Munsell, Robert S. Harris and Louis O. Williams** (*by invitation*), with the technical assistance of **Louise Guild** (*by invitation*), **Gertrude Nightingale** (*by invitation*) **and Cynthia Troescher** (*by invitation*), *Nutritional Biochemistry Laboratories, Massachusetts Institute of Technology*

Composition of Central American foods: I. Honduras.

9. **Leon M. Sharpe** (*by invitation*), **Robert S. Harris, Wendell C. Peacock** (*by invitation*) **and Richard C. Cooke** (*by invitation*), *Dept. of Food Technology and Physics, Massachusetts Institute of Technology and the Walter E. Fernald State School*

Effect of phytate and other food ingredients on the absorption of radioactive iron.

10. **Nathan W. Shock and Marvin J. Yiengst** (*by invitation*), *Division of Physiology, National Institute of Health and Baltimore City Hospitals*

Experimental displacement of the acid-base equilibrium of the blood in aged males.

PHYSIOLOGY A

Thursday, March 18, 1:45 p.m.

Ballroom, Convention Hall

SYMPOSIUM

Physiology of Neuro-Muscular Junctions

John H. Welsh, *Chairman*

1. **John H. Welsh,** *Harvard University*

Introduction.

2. **Stephen W. Kuffler,** *Johns Hopkins University*

Electrical Aspects.

3. **George H. Acheson,** *Harvard University*

Chemical Aspects.

4. **A. McGehee Harvey,** *Johns Hopkins Univ.*

Clinical Aspects.

PHYSIOLOGY B

Thursday, March 18, 1:45 p.m.

Room 20, Convention Hall

Cardiac Work

1. **Doris J. W. Escher** (*by invitation*), **Raymond E. Weston, George Leiner** (*by invitation*), **Louis Leiter** (*by invitation*) **and Sybil Goldat** (*by invitation*), *Medical Division, Montefiore Hospital, New York*

The effect of aminophyllin on cardiac output and renal hemodynamics in man.

2. **Joseph R. DiPalma and Richard A. Reiss** (*by invitation*), *Dept. of Physiology, Long Island College of Medicine*

The mechanics of Starling's law of the heart.

3. **Richard A. Reiss** (*by invitation*) **and Joseph R. DiPalma,** *Dept. of Physiology, Long Island College of Medicine*
   The effect of changes in cardiac "competence" on right auricular pressure.
4. **J. E. Eckenhoff, J. H. Hafkenschiel, E. L. Foltz and R. L. Driver** (*introduced by* Carl F. Schmidt), *Dept. of Pharmacology and Harrison Dept. of Surgical Research, School of Medicine, University of Pennsylvania*
   The influence of hypotension on coronary blood flow, cardiac oxygen metabolism and cardiac work.
5. **Albert Roos** (*by invitation*), **J. O. Elam** (*by invitation*), **J. F. Neville, Jr.** (*by invitation*) **and H. L. White,** *Dept. of Physiology and Laboratory of Applied Thoracic Physiology, Washington University School of Medicine*
   Oximetric determination of cardiac output in man.
6. **W. F. Hamilton, R. L. Riley** (*by invitation*), **A. M. Attyah** (*by invitation*), **Andre Cournand, D. M. Fowell** (*by invitation*), **A. Himmelstein** (*by invitation*), **R. P. Noble** (*by invitation*), **J. W. Remington, D. W. Richards, Jr.** (*by invitation*), **N. C. Wheeler** (*by invitation*) **and A. C. Witham** (*by invitation*), *Dept. of Physiology, University of Georgia School of Medicine, and the Chest Service, Bellevue Hospital, Columbia University*
   Comparison of the Fick and dye injection methods of measuring the cardiac output in man.
7. **D. M. Fowell** (*by invitation*), **A. P. Briggs, N. C. Wheeler** (*by invitation*), **J. A. Winslow, Jr.** (*by invitation*), **J. W. Remington and W. F. Hamilton,** *Depts. of Physiology, Biochemistry and Medicine, University of Georgia School of Medicine*
   Renal and circulatory factors in congestive failure of the circulation.
8. **John W. Remington, W. F. Hamilton, Charles R. Noback** (*by invitation*) **and Jay J. Gold** (*by invitation*), *Dept. of Physiology, University of Georgia School of Medicine and Dept. of Anatomy, Long Island Medical School*
   Prediction of the stroke volume of man from brachial pressure pulse values.
9. **Orville Horwitz** (*by invitation*), **Robert L. Mayock** (*by invitation*) **and Isaac Starr,** *Dept. of Therapeutic Research and the Dept. of Medicine, University of Pennsylvania*
   Direct experiments on the relation between the form of the ballistocardiogram and the shape of the systolic velocity curve in the aorta of man.
10. **J. C. Handelsman** (*by invitation*), **Richard J. Bing and L. D. Vandam** (*by invitation*), *Dept. of Surgery, Johns Hopkins University and Hospital*
    Physiological adaptations to anoxia in congenital heart disease with cyanosis.
11. **Gordon C. Ring, Catharine R. Michie** (*by invitation*) **and M. J. Oppenheimer,** *Dept. of Physiology, Temple University School of Medicine*
    X-ray density changes in the heart recorded by the electrokymograph.
12. **Edward F. Randak** (*by invitation*), **Bert R. Boone** (*by invitation*), **George F. Ellinger** (*by invitation*) **and M. J. Oppenheimer,** *Dept. of Physiology, Temple University School of Medicine and Heart Disease Control Section, U. S. Public Health Service*
    Ventricular isometric relaxation phase as measured on the electrokymogram.

PHYSIOLOGY BUSINESS MEETING

Thursday, March 18, 4:15 p.m.

ROOM 20, CONVENTION HALL

PHYSIOLOGY C

Thursday, March 18, 1:45 p.m.

ROOM 21, CONVENTION HALL

Gastro-Intestinal Functions

1. **R. F. Furchgott** (*by invitation*) **and Ephraim Shorr,** *Dept. of Medicine, Cornell University Medical College and The New York Hospital*
   Effect of anoxia on contractility and metabolism of intestinal smooth muscle.
2. **Emil Bozler,** *Dept. of Physiology, Ohio State University*
   The myenteric reflex.
3. **Daniel A. Brody** (*by invitation*) **and J. P. Quigley,** *Dept. of Physiology, University of Tennessee*
   Measurement of gastric tonus in the normal human.
4. **Kao Hwang** (*introduced by* A. C. Ivy), *Dept. of Clinical Science, University of Illinois College of Medicine*
   Reflex activities and peristaltic function of the esophagus.
5. **David W. Northup, J. C. Stickney and E. J. Van Liere,** *West Virginia University Medical School*
   The effect of carbon dioxide on the motility of the small intestine.
6. **K. L. Shapiro** (*by invitation*) **and A. C. Ivy,** *Dept. of Clinical Science, University of Illinois College of Medicine*

The effect of "carbonation" on the rate of gastric emptying and intestinal absorption of sucrose solution.

7. **Dean A. Collins,** *Dept. of Physiology, Temple University School of Medicine*
   Effect of tetraethylammonium on responses of isolated intestine to angiotonin and other substances.
8. **G. M. Morton** (*by invitation*) **and G. W. Stavraky,** *Depts. of Anatomy and Physiology, Faculty of Medicine, University of Western Ontario*
   The effect of intra-arterial injection of acetylcholine upon the gastric mucosa of the dog.
9. **C. C. Wang** (*by invitation*) **and M. I. Grossman,** *Dept. of Clinical Science, University of Illinois College of Medicine*
   Effect of secretin and pancreozymin on amylase and alkaline phosphatase of dog's pancreas.
10. **I. J. Pincus** (*by invitation*), **J. E. Thomas and P. O. Lachman** (*by invitation*), *Jefferson Medical College*
    The effect of vagotomy on the secretion of pancreatic juice after the ingestion of various foodstuffs.
11. **Robert E. Davis** (*by invitation*) **and A. C. Ivy,** *Dept. of Clinical Science, University of Illinois College of Medicine*
    Thermal irritation of alimentary mucosa.
12. **W. Barrett** (*by invitation*) **and B. Craver,** *Division of Pharmacology, Research Dept., Ciba Pharmaceutical Products, Inc.*
    Effect of ion-deficient Locke's solution on guinea pig ileal responses to histamine, acetylcholine, electrical stimulation. (Pharmacol.)
13. **C. J. Podore** (*by invitation*), **R. H. Broh-Kahn** (*by invitation*) **and I. Arthur Mirsky,** *May Institute for Medical Research of the Jewish Hospital, Cincinnati*
    Uropepsin excretion in man.

PHYSIOLOGY BUSINESS MEETING

Thursday, March 18, 4:15 p.m.

Room 20, Convention Hall

PHYSIOLOGY D

Thursday, March 18, 1:45 p.m.

Room 22, Convention Hall

Liver Physiology

1. **Attilio Canzanelli, David Rapport and Ruth Guild** (*by invitation*), *Dept. of Physiology, Tufts College Medical School*
   Effect of diet on the in vitro synthesis of alamine by liver.
2. **Carl E. Anderson** (*by invitation*), **Robert G. Gale** (*by invitation*) **and C. S. Robinson,** *Depts. of Biochemistry and Medicine, Vanderbilt University*
   The estimation of functional hepatic mass in normal and phosphorus-poisoned dogs. (Biochem.)
3. **R. W. Denton** (*by invitation*) **and A. C. Ivy,** *Dept. of Clinical Science, University of Illinois College of Medicine*
   Liver regeneration in the rat.
4. **Edward A. Newman** (*by invitation*) **and Morton I. Grossman,** *Dept. of Clinical Science, University of Illinois College of Medicine*
   The effect of diet on liver regeneration in partially hepatectomized rats.
5. **Clara M. Szego and Sidney Roberts,** *Dept. of Physiological Chemistry, Yale University*
   The chemical composition of regenerating rat liver: the influence of ovariectomy.
6. **Ephraim Shorr and B. W. Zweifach,** *Dept. of Medicine, Cornell University Medical College and The New York Hospital*
   Hepato-renal factors in circulatory homeostasis. XIX. VEM and VDM mechanisms in nutritional cirrhosis in rats.
7. **Stephen Leslie** (*by invitation*), **George H. Stueck, Jr.** (*by invitation*), **Harold M. Shorr** (*by invitation*) **and Elaine P. Ralli,** *Dept. of Medicine, New York University College of Medicine*
   Chemical and physiological changes during water tolerance tests in patients with cirrhosis.
8. **J. F. Canepa** (*by invitation*) **and A. C. Ivy,** *Dept. of Clinical Science, University of Illinois College of Medicine*
   Comparative lipotropic activity of parenterally administered pancreatic extracts in dietary fatty livers.
9. **A. E. Groff** (*by invitation*), **E. deF. Baldwin** (*by invitation*) **and S. E. Bradley,** *Dept. of Medicine, College of Physicians and Surgeons, Columbia University and the Presbyterian Hospital, New York*
   Hepatic arterio-venous oxygen differences in patients with normal and diseased livers.
10. **Philip K. Bondy, Walter H. Sheldon and Lillian Evans** (*introduced by* James V. Warren), *Depts. of Medicine and Pathology, Emory University School of Medicine*
    Quantitative glycogen determinations on specimens of human livers obtained by needle biopsy.
11. **N. R. Brewer** (*introduced by* A. B. Luckhardt) **and R. L. Gunning,** *Dept. of Physiology, University of Chicago*

The secretory pressure of the liver of the chicken.

12. **W. J. Snape** (*by invitation*), **C. W. Wirts** (*by invitation*), **L. L. Miller and A. Cantarow,** *Jefferson Medical College*
Excretion of bilirubin and bromsulfalein by the liver.

PHYSIOLOGY BUSINESS MEETING

Thursday, March 18, 4:15 p.m.

ROOM 20, CONVENTION HALL

PHYSIOLOGY E

Thursday, March 18, 1:45 p.m.

ROOM 17, CONVENTION HALL

Brain Physiology

1. **E. A. Spiegel and H. T. Wycis** (*by invitation*), *Dept. of Experimental Neurology, Temple University School of Medicine*
Electroencephalographic studies following thalamic lesions in humans.
2. **F. E. Nulsen, S. P. W. Black and C. V. Drake** (*by invitation*), *Laboratory of Physiology, Yale University School of Medicine*
Inhibition and facilitation of motor activity by the anterior cerebellum.
3. **Ray S. Snider** (*by invitation*) **and H. W. Magoun,** *Dept. of Anatomy, Northwestern University School of Medicine*
Facilitation produced by cerebellar stimulation.
4. **Joseph Thomas Roberts,** *University of Arkansas School of Medicine and Gallinger Municipal Hospital, Washington*
The role of the *vasa nervorum*, especially in regard to "referred pain".
5. **F. H. Lewey,** *Peripheral Nerve Study Center, Hospital of the University of Pennsylvania*
Quantitative examination of sensibility in peripheral nerve injuries.
6. **L. W. Freeman, H. B. Shumacker, Jr.** (*by invitation*), **E. E. Wayson** (*by invitation*) **and N. M. Stahl** (*by invitation*), *Dept. of Surgery, Yale University School of Medicine*
Conduction of painful impulses from the extremities via the sympathetic nervous system.
7. **James D. Hardy and Carl T. Javert** (*by invitation*), *Russell Sage Institute of Pathology, The New York Hospital, and the Depts. of Physiology, Gynecology and Obstetrics, Cornell University Medical College*
Experimental investigation of pain intensity in labor.
8. **Abraham Wiker and Karl Frank** (*by invitation*), *Research Dept., U. S. Public Health Hospital, Lexington, Ky.*
Effects of electroshock convulsions on chronic decorticated cats. (Pharmacol.)
9. **John M. Brookhart and Russell E. Morris** (*by invitation*), *Institute of Neurology, Northwestern University Medical School*
Antidromic potential recordings from the medullary pyramid of the cat.
10. **L. M. N. Bach** (*introduced by* H. S. Mayerson), *Dept. of Physiology, Tulane University School of Medicine*
The role of the bulbar facilitatory and inhibitory systems in vasomotor and respiratory activity.
11. **W. Horsley Gantt,** *Pavlovian Laboratory of the Phipps Psychiatric Clinic, Johns Hopkins Hospital*
Effect of satiation on the intensity of the conditional and unconditional salivary secretion.
12. **J. M. Essenberg** (*introduced by* George Clark), *Dept. of Anatomy, The Chicago Medical School*
The effect of nicotine on maze-learning ability of albino rats.

PHYSIOLOGY BUSINESS MEETING

Thursday, March 18, 4:15 p.m.

ROOM 20, CONVENTION HALL

PHYSIOLOGY

Motion Pictures

Thursday, March 18, 8:00 p.m.

ROOM C, CONVENTION HALL

Program on page 2

PHYSIOLOGY A

Friday, March 19, 9:00 a.m.

BALLROOM, CONVENTION HALL

Renal Mechanisms

1. **L. J. Cizek** (*by invitation*) **and J. H. Holmes,** *Dept. of Physiology, College of Physicians and Surgeons, Columbia University*
Studies of the mechanism of increased chloride excretion during osmotic diuresis.
2. **Laurence G. Wesson, W. Parker Anslow, Jr. and Homer W. Smith,** *New York University College of Medicine*
The renal excretion of strong electrolytes.
3. **J. C. Roemmelt** (*by invitation*), **O. W. Sartorius** (*by invitation*) **and R. F. Pitts,**

Dept. of Physiology, Syracuse University College of Medicine

Sodium and chloride reabsorption.

4. **R. F. Pitts, J. L. Ayer and W. A. Schiess,** *Dept. of Physiology, Syracuse University College of Medicine*

The reabsorption and excretion of bicarbonate in normal man.

5. **Iftakhar Jahan** (*by invitation*) **and R. F. Pitts,** *Dept. of Physiology, Syracuse University College of Medicine*

The effect of parathormone on the renal tubular reabsorption of inorganic phosphate.

6. **W. D. Lotspeich** (*introduced by* R. F. Pitts), *Dept. of Physiology, Syracuse University College of Medicine*

Glomerular filtration rate in the adrenalectomized, salt-fed rat.

7. **O. W. Sartorius** (*by invitation*), **J. C. Roemmelt** (*by invitation*) **and R. F. Pitts,** *Dept. of Physiology, Syracuse University College of Medicine*

Changes in renal function in experimental metabolic acidosis in the normal human subject.

8. **W. Parker Anslow, Jr., Laurence G. Wesson, Jr., Alfred A. Bolomey and John G. Taylor** (*introduced by* Homer W. Smith), *New York University College of Medicine*

Chloruretic action of pressor-antidiuretic fraction of posterior pituitary extract.

9. **Elizabeth O'Leary** (*by invitation*) **and Samuel A. Corson,** with the technical assistance of Opal Cain, Catherine Anderson and Elinor Foster, *Dept. of Physiology, School of Medicine, University of Minnesota and Howard University*

Renal clearance studies in experimental hypoproteinemic edema.

10. **W. Q. Wolfson** (*by invitation*) **and R. Levine,** *Depts. of Biochemistry and Metabolic and Endocrine Research, Medical Research Institute, Michael Reese Hospital, Chicago*

The transport and excretion of uric acid in man. IV. The renal mechanism for urate excretion.

11. **Henry L. Barnett** (*by invitation*), **Helen McNamara** (*by invitation*), **Ruth S. Hare** (*by invitation*) **and Kendrick Hare,** *New York Hospital and Dept. of Pediatrics, Cornell University Medical College*

Inulin, urea, mannitol and PAH clearance ratios in premature infants.

12. **C. Boyd Shaffer, Frances H. Critchfield and Charles P. Carpenter** (*introduced by* J. M. Rogoff), *Chemical Hygiene Fellowship, Mellon Institute, Pittsburgh.*

The renal clearance of some polyethylene glycols in the dog.

PHYSIOLOGY B

Friday, March 19, 9:00 a.m.

Room 20, Convention Hall

Peripheral Circulation

1. **Clifford G. Gaddy** (*by invitation*), **Harold D. Green and J. Maxwell Little,** *Dept. of Physiology and Pharmacology, Bowman Gray School of Medicine, Wake Forest College*

Peripheral vasodilator effect of a substance present in normal human urine.

2. **Vivian G. Behrmann** (*introduced by* Robert Gesell), *Dept. of Laboratories, Henry Ford Hospital*

Oxyhemograph studies during marked vasomotor changes.

3. **A. B. Hertzman and W. C. Randall,** *Dept. of Physiology, St. Louis University Medical School*

Further studies on the correlation between skin volume pulses and blood flow.

4. **K. G. Wakim, J. C. Terrier** (*by invitation*) **and E. C. Elkins** (*by invitation*), *Mayo Clinic*

The effects of percutaneous stimulation on the circulation in normal and in paralyzed extremities.

5. **G. A. Hallenbeck, E. H. Wood and O. T. Clagett** (*by invitation*), *Section on Physiology and Division of Surgery, Mayo Clinic*

Changes in radial arterial blood pressure during surgical resection of coarctation of the aorta.

6. **F. Olmsted** (*by invitation*), **A. C. Corcoran, O. Glasser** (*by invitation*) **and Irvine H. Page,** *Research Division of the Cleveland Clinic Foundation*

Systolic pressure in the intact unanesthetized rat.

7. **R. Lenel** (*introduced by* S. Rodbard), *Cardiovascular Dept., Research Institute, Michael Reese Hospital, Chicago*

The secondary blood pressure rise and tachycardia occurring after the injection of epinephrine.

8. **D. M. Green, A. D. Johnson, A. Lobb and G. Cusick** (*introduced by* R. Frederick Becker), *University of Washington School of Medicine*

The relation of initial blood pressure to adrenalin action.

9. **Kenneth E. Jochim,** *Dept. of Physiology, University of Kansas*

Arterial pulses simulated in electrical analogues of the circulatory system.

10. **Wilson C. Grant** (*introduced by* Walter S. Root), *Dept. of Physiology, College of Physicians and Surgeons, Columbia University*

Oxygen in bone marrow blood during prolonged hemorrhagic anemia.

11. **Hardin B. Jones** (*introduced by* Nello Pace), *Division of Medical Physics, University of California*
Changes in tissue vascularity and inert gas exchange related to age.
12. **V. Winder and Mona M. Anderson** (*by invitation*), *Research Laboratories, Parke, Davis & Co., Detroit*
On the nature of tachyphylaxis and related phenomena.

PHYSIOLOGY C

Friday, March 19, 9:00 a.m.

ROOM 21, CONVENTION HALL

Adrenals

1. **Joseph T. King, Carmen B. Casas** (*by invitation*) **and Claire J. Carr** (*by invitation*), *Dept. of Physiology, University of Minnesota*
Effect of corticotropin on ovariectomized C3H mice bearing adrenal adenomas.
2. **Leon Hellman** (*by invitation*), **Raymond E. Weston, Doris J. W. Escher** (*by invitation*) **and Louis Leiter** (*by invitation*), *Medical Division, Montefiore Hospital, New York*
The effect of adrenocorticotropin on renal hemodynamics and uric acid clearance.
3. **Oscar Hechter and David Stone** (*by invitation*), *Worcester Foundation for Experimental Biology and Dept. of Physiology, Tufts College Medical School*
In vitro action of adrenal cortical extract upon lymphocytes.
4. **D. J. Ingle and M. C. Prestrud** (*by invitation*), *Research Laboratories, The Upjohn Company, Kalamazoo*
Changes in urinary glucose and nitrogen following adrenalectomy in the force-fed diabetic rat.
5. **K. E. Paschkis, A. Cantarow, A. A. Walkling** (*by invitation*), **W. H. Pearlman, A. E. Rakoff** (*by invitation*) **and D. Boyle** (*by invitation*), *Jefferson Medical College*
Secretion and excretion of carbohydrate-active adrenal compounds (oxysteroids)
6. **Gregory Pincus, Louise Romanoff** (*by invitation*) **and James Carlo** (*by invitation*), *Worcester Foundation for Experimental Biology*
Variations with age in neutral steroid excretion of men.
7. **Hans Seyle, Pierre Haour** (*by invitation*) **and Claude Faribault** (*by invitation*), *Institut de Medecine et de Chirurgie Experimentales, Universite de Montreal*
Further studies concerning brain lesions induced by desoxycorticosterone overdosage in the rat.
8. **Hudson Hoagland and David Stone** (*by invitation*), *Worcester Foundation for Experimental Biology*
Brain and muscle potassium in relation to stressful activities and adrenal cortical function.
9. **Marguerite N. Swift, Harvey M. Patt and Ella B. Tyree** (*introduced by* Austin M. Brues), *Biology Division, Argonne National Laboratory*
The effect of adrenal cortical extract on adrenal response to total body X-irradiation.
10. **A. Van Loo** (*by invitation*), **A. Surtshin** (*by invitation*) **and L. N. Katz,** *Cardiovascular Dept., Research Institute, Michael Reese Hospital, Chicago*
The role of the adrenal in the arterial pressure responses to severe hypoxemia.
11. **L. Recant** (*by invitation*), **P. H. Forsham** (*by invitation*) **and G. W. Thorn,** *Dept. of Medicine, Harvard Medical School, and the Medical Clinic, Peter Bent Brigham Hospital*
Effect of epinephrine on the circulating eosinophils.

PHYSIOLOGY D

Friday, March 19, 9:00 a.m.

ROOM 22, CONVENTION HALL

Special Senses

1. **C. Haig and E. M. Haig** (*by invitation*), *Dept. of Physiology and Biochemistry, New York Medical College*
The brightness threshold as a function of area and receptor number in various retinal regions.
2. **E. F. MacNichol** (*by invitation*) **and H. K. Hartline,** *Johnson Research Foundation, University of Pennsylvania*
Responses to small changes of light intensity by the light-adapted photoreceptor.
3. **Hans-Lukas Teuber** (*by invitation*) **and Morris B. Bender,** *Psychophysiological Laboratory, Dept. of Neurology, New York University College of Medicine*
Critical flicker frequency in defective fields of vision.
4. **George M. Austin** (*by invitation*), **F. H. Lewey and Francis C. Grant** (*by invitation*), *Laboratory of the Neurosurgical Service, Hospital of the University of Pennsylvania*
Physiological significance of preserved central vision in lesions of the optic tract vs. optic radiation.
5. **George Wald,** *Biological Laboratories, Harvard University*
Interconversions of retinene and vitamin A in vitro.

6. **Charles Sheard,** *Division of Physics and Biophysical Research, Mayo Foundation and Mayo Clinic*
   Courses of dark adaptation and levels of vitamin A and carotene in normal and clinical conditions.
7. **Stanley K. Davis** (*introduced by* J. W. Heim), *Aero Medical Laboratory, Wright Field*
   Influence of intravenous cytochrome C upon visual acuity of the dark adapted human eye.
8. **William M. Hart and Bruce F. Chandler** (*by invitation*), *Depts. of Ophthalmology and Biochemistry, Temple University School of Medicine*
   Factors responsible for transmission of visible light by the fibrous tunic of the eye.
9. **J. L. Patterson, Jr. and Ashton Graybiel** (*introduced by* James V. Warren), *Naval School of Aviation Medicine and Research, Pensacola, and Dept. of Physiology, Emory University Medical School*
   A negative form of the oculo-gravic illusion.
10. **Irving H. Wagman,** *Dept. of Physiology, Jefferson Medical College*
    Pupil dilatation in darkness as effected by intensity and duration of pre-exposure to white light.
11. **Freya Stone** (*by invitation*) **and Franz R. Goetzl,** *Dept. of Medical Research, Permanente Foundation*
    Olfactory acuity and appetite: effects of bitter tonics on olfactory acuity in normal human subjects.
12. **Elwood Henneman and Vernon Mountcastle** (*introduced by* Philip Bard), *Dept. of Physiology, Johns Hopkins University School of Medicine*
    Tactile localization in the thalamus of cat and monkey.

PHYSIOLOGY E

Friday, March 19, 9:00 a.m.

ROOM 17, CONVENTION HALL

Muscle

1. **Ernst Fischer and Russel V. Bowers** (*by invitation*), *Baruch Center of Physical Medicine, Medical College of Virginia*
   Changes in zymohexase activity during denervation atrophy of skeletal muscle and their retardation by appropriate electrical treatment.
2. **William J. Bowen and William E. Poel** (*introduced by* Heinz Specht), *Laboratory of Physical Biology, National Institute of Health, Bethesda*
   The effects of anoxia upon myoglobin concentration.
3. **William R. Amberson, Rubert S. Anderson** (*by invitation*), **Betty Chinn** (*by invitation*) **and T. Erdös** (*by invitation*), *Dept. of Physiology, University of Maryland, and Institute of Physical Chemistry, University of Uppsala*
   Extraction of myogen and myosin from whole skeletal muscles.
4. **Julian M. Tobias,** *Dept. of Physiology and Toxicity Laboratory, University of Chicago*
   Injury potential in potassium-depleted frog muscle.
5. **Sheppard M. Walker,** *Dept. of Physiology, Washington University School of Medicine*
   Action potentials induced by indirect stimulation of rat muscle after adrenalectomy, KCl treatment and tetanus.
6. **J. S. Denslow and David M. Graham-Service** (*introduced by* Irvin M. Korr), *Still Memorial Research Trust, Kirksville*
   The spread of muscle action potentials from active to inactive areas.
7. **G. Ling** (*introduced by* R. W. Gerard), *Dept. of Physiology, University of Chicago*
   Effect of stretch on membrane potential in frog muscle.
8. **Alexander Sandow,** *Washington Square College of Arts and Sciences, New York University*
   Transverse latency relaxations of muscle stimulated with massive transverse shocks.
9. **H. J. Ralston, J. R. Close** (*by invitation*), **V. T. Inman** (*by invitation*) **and B. Feinstein** (*by invitation*), *Dept. of Physiology, College of Physicians and Surgeons, San Francisco, and the University of California School of Medicine and College of Engineering*
   Dynamical and electrical features of human isolated voluntary muscle in isometric and isotonic contraction.
10. **Arthur W. Martin and O. M. Sola** (*by invitation*), *Dept. of Physiology and Biophysics, University of Washington*
    The rate of atrophy of rat diaphragm.
11. **Ernest G. Huf** (*introduced by* Ernst Fischer), *Baruch Center of Physical Medicine and Dept. of Physiology, Medical College of Virginia*
    Muscular atrophy as a state of "local hyperthyroidism."

PHYSIOLOGY A

Friday, March 19, 1:45 p.m.

BALLROOM, CONVENTION HALL

Cerebral Cortex

1. **Ernest Sachs, Jr. and Samuel Brendler** (*introduced by* John F. Fulton), *Laboratory*

of Physiology, Yale University School of Medicine

Some effects of stimulation of the orbital surface of the frontal lobe in the dog and monkey.

2. **Robert B. Livingston** (*by invitation*) **and John F. Fulton,** *Laboratory of Physiology, Yale University School of Medicine*

"Cortical instabiity": a study of frequency effects.

3. **Wilder Penfield and Keasley Welch** (*by invitation*)

Instability of motor points and sensory points in the human cerebral cortex.

4. **W. S. McCulloch and Elwood Henneman,** *Dept. of Psychiatry, University of Illinois*

The projection of area 19 to the reticular formation.

5. **Clinton N. Woolsey and D. H. LeMessurier** (*by invitation*), *Dept. of Physiology, Johns Hopkins University School of Medicine*

The pattern of cutaneous representation in the rat's cerebral cortex.

6. **Fred A. Mettler and J. Lawrence Pool** (*by invitation*), *Dept. of Neurology, College of Physicians and Surgeons, Columbia University*

Removal of restral border of human area 4 followed by spasticity or lack of it.

7. **J. E. Ziegler and T. W. Rasmussen** (*introduced by* Herbert Jasper), *Dept. of Neurology and Neurosurgery, McGill University, and the Montreal Neurological Institute*

The effect of local graded pressure upon electrical activity and excitability of the motor cortex.

8. **Richard G. Berry and Francis M. Forster** (*introduced by* M. H. F. Friedman), *Dept. of Neurology, Jefferson Medical College*

Cerebral cortical effects of curare.

9. **John L. Hampson** (*introduced by* C. N. Woolsey), *Dept. of Physiology, Johns Hopkins University School of Medicine*

Relationships between the cerebral and cerebellar cortices in the cat.

10. **Herbert H. Jasper and Jan Droogleever-Fortuyn** (*by invitation*), *Dept. of Neurology and Neurosurgery, McGill University and the Montreal Neurological Institute*

Thalamo-cortical systems and the electrical activity of the brain.

11. **Eli S. Goldensohn** (*by invitation*), **Ewald W. Dusse** (*by invitation*), **Joseph N. Spencer** (*by invitation*), **William B. Draper and Richard W. Whitehead,** *Depts. of Physiology and Pharmacology, and Psychiatry, University of Colorado Medical Center*

Studies on diffusion respiration. VII. Electrical cortical activity in dogs. (Pharmacol.)

12. **Charles D. Hendley** (*introduced by* Horace W. Davenport), *Depts. of Physiology and Pharmacology, University of Utah School of Medicine*

The effect of blood acid-base changes on convulsive seizures.

PHYSIOLOGY B

Friday, March 19, 1:45 p.m.

Room 20, Convention Hall

Biophysics

1. **F. H. Johnson, M. B. Baylor** (*by invitation*) **and D. Fraser** (*by invitation*), *Dept. of Biology, Princeton University*

The thermal denaturation of tobacco mosaic virus in relation to hydrostatic pressure.

2. **Harvey M. Patt, Marguerite N. Swift and Ella B. Tyree** (*introduced by* Austin M. Brues), *Biology Division, Argonne National Laboratory*

Influence of temperature on radio-sensitivity in the frog (rana pipiens).

3. **H. O. Parrack and D. H. Eldredge, Jr.** (*introduced by* H. M. Sweeney), *Aero Medical Laboratory, Air Materiel Command, Wright Field*

Certain physiological reactions to intense sound fields.

4. **Alice M. Stoll** (*by invitation*), **and James D. Hardy,** *Russell Sage Institute of Pathology, The New York Hospital, and the Dept. of Physiology, Cornell University Medical College*

Direct experimental comparison of several surface temperature-measuring devices.

5. **Charles H. Richards** (*by invitation*) **and James D. Hardy,** *The New York Hospital and the Dept. of Physiology, Cornell University Medical College*

An instrument for the measurement of total thermal radiation in the environment.

6. **Ronald Deering** (*introduced by* A. B. Hertzman), *Dept. of Physiology, St. Louis University Medical School*

The rod calorimeter.

7. **E. E. Painter and M. C. Moore** (*by invitation*), *Loyola University School of Medicine, Argonne National Laboratory, and University of Illinois College of Medicine*

Susceptibility of X-rayed animals to histamine and to adenosine.

8. **Herbert Shapiro,** *Laboratory of Physical Biology, National Institute of Health, Bethesda*

The alteration in osmotically inactive fraction produced by cell activation.

9. **Richard McFee** (*introduced by* J. S. Robb), *Dept. of Pharmacology, Syracuse University College of Medicine*
   A critical discussion of the bioelectric "doublet" theory.
10. **David F. Waugh, M. Janette Smith** (*by invitation*) **and Darthea F. Fearing** (*by invitation*), *Dept. of Biology, Massachusetts Institute of Technology*
    Regeneration of insulin fibrils with several reagents and the nature of the inter-insulin bond.

PHYSIOLOGY C

Friday, March 19, 1:45 p.m.

ROOM 21, CONVENTION HALL

Circulation and Heart

1. **Robert W. Ramsey, George Fischer** (*by invitation*), **and Marie Louise Flinker** (*by invitation*), *Depts. of Physiology and Pharmacology, Medical College of Virginia*
   The effect of eserine on the absolutely and relatively refractory period of turtle heart strips.
2. **Frank L. Pettinga** (*by invitation*) **and J. W. Stutzman,** *Dept. of Pharmacology, Boston University School of Medicine*
   Effect of adrenalectomy or evisceration on cyclopropane-induced cardiac arrhythmias in cats.
3. **Ingrith J. Deyrup** (*by invitation*) **and William W. Walcott,** *Dept. of Physiology, College of Physicians and Surgeons, Columbia University*
   A study of the effects of the intravenous injection of hypertonic solutions on the heart rates of cats and dogs.
4. **Augustus C. P. Bakos** (*introduced by* Charles F. Morgan), *Dept. of Physiology and Biophysics, Georgetown University School of Medicine*
   The maintenance of mean pulmonary arterial pressure after extreme right ventricular damage in the dog. (Pharmacol.)
5. **J. Richard R. Bobb, Donald C. Kunze and William McCall, Jr.** (*introduced by* Harold D. Green), *Dept. of Physiology and Pharmacology, Bowman Gray School of Medicine of Wake Forest College*
   A method for production of cardiac infarction in the dog.
6. **L. H. Peterson** (*introduced by* H. C. Bazett) **and G. C. Risman** (*introduced by* H. C. Bazett), *Dept. of Physiology, School of Medicine, University of Pennsylvania*
   Direct blood pressure recording in man.
7. **John R. Smith and Masauki Hara** (*by invitation*), *Dept. of Internal Medicine, Washington University School of Medicine*
   Experimental embolism of selected portions of pulmonary arterial bed.
8. **Maurice B. Visscher,** *University of Minnesota*
   The capacity changes in the pulmonary vascular bed with the respiratory cycle.
9. **R. L. Riley** (*by invitation*), **A. Himmelstein** (*by invitation*), **H. L. Motley** (*by invitation*), **H. M. Weiner** (*by invitation*) **and A. Cournand,** *Cardio-Pulmonary Laboratory, Chest Service, Bellevue Hospital, and the Dept. of Medicine, College of Physicians and Surgeons, Columbia University*
   Pulmonary circulation during exercise in normal individuals and in patients with chronic pulmonary disease.
10. **Joseph H. Hafkenschiel and James E. Eckenhoff** (*introduced by* Carl F. Schmidt), *Dept. of Pharmacology and Harrison Dept. of Surgical Research, School of Medicine, University of Pennsylvania*
    The oxygen content of coronary venous blood as affected by anoxia and cytochrome C. (Pharmacol.)
11. **Cecil K. Drinker and Esther Hardenbergh** (*by invitation*), *Dept. of Physiology, Harvard School of Public Health*
    Acute effects upon the lungs of dogs of large intravenous doses of alpha-naphythyl thiourea (ANTU).
12. **Harold Koenig** (*introduced by* W. F. Windle) **and Ruth Koenig** (*introduced by* W. F. Windle). *Anatomical Laboratories, University of Pennsylvania School of Medicine*
    Acute pulmonary edema produced by ammonium salts.

PHYSIOLOGY D

Friday, March 19, 1:45 p.m.

ROOM 22, CONVENTION HALL

Metabolism

1. **Francis X. Fellers** (*by invitation*), **Henry L. Barnett** (*by invitation*), **Helen McNamara** (*by invitation*) **and Kendrick Hare,** *The New York Hospital and Dept. of Pediatrics, Cornell University Medical College*
   Decrease in the radiosodium and thiocyanate spaces during growth.
2. **K. I. Altman, G. W. Casarett, R. E. Masters, T. R. Noonan, and K. Salomon,** (*introduced by* W. F. Bale), *University of Rochester School of Medicine and Dentistry, Atomic Energy Project*
   Hemin synthesis with glycine containing C^{14} in its alpha-carbon atom.
3. **John M. Anderson,** (*introduced by* Otto M. Helff), *Dept. of Biology, Brown University*

Changes in nitrogen distribution in the Japanese beetle during metamorphosis.

4. **R. Levine and N. G. Schneeberg** (*by invitation*), *Dept. of Metabolism and Endocrinology, Research Institute, Michael Reese Hospital, Chicago*

The disposal of intravenously administered amino acids by normal, hepatectomized, depancreatized and hyperthyroid dogs.

5. **C. Martin Rhode** (*by invitation*), **William M. Parkins and Harry M. Vars,** *Harrison Dept. of Surgical Research, Schools of Medicine, Univ. of Pennsylvania*

Nitrogen balances of dogs continuously infused with 50% glucose and protein preparations.

6. **L. Van Middlesworth and D. H. Copp** (*introduced by* J. P. Quigley), *Division of Physiology, University of California, and University of Tennessee*

Post-fracture nitrogen loss with and without methionine supplements to good and poor protein diets.

7. **Jay A. Smith** (*by invitation*), **Piero P. Foa, Harriet R. Weinstein** (*by invitation*), **A. Sidney Ludwig** (*by invitation*), **and J. Marvin Wertheim** (*by invitation*), *Dept. of Physiology and Pharmacology, Chicago Medical School*

Some toxic effects of acid and neutralized thiamine solutions.

8. **Isaac M. Berry** (*by invitation*) **and A. C. Ivy,** *Dept. of Clinical Science, University of Illinois College of Medicine*

The tolerance of dogs to intravenously administered fatty chyle and synthetic fat emulsion.

9. **Piero P. Foa and Harriet R. Weinstein** (*by invitation*), *Dept. of Physiology and Pharmacology, Chicago Medical School*

The lipids of the rat brain in choline deficiency.

10. **M. H. Hack** (*introduced by* Hubert R. Catchpole), *Dept. of Pathology, University of Illinois College of Medicine*

The phosphatide composition of human erythrocytes.

11. **Wiktor W. Nowinski** (*introduced by* C. M. Fomerat), *Tissue Culture Laboratory and the Psychopathic Hospital, Medical Branch, University of Texas*

Influence of anti-organ sera upon the oxygen uptake of the spleen and brain.

12. **J. H. Peters** (*by invitation*), **L. Greenman** (*by invitation*), **and T. S. Danowski,** *Dept. of Research Medicine and the Renziehausen Foundation University of Pittsburgh School of Medicine*

Beneficial effects of calcium chloride in fluoride poisoning.

PHYSIOLOGY E

Friday, March 19, 1:45 p.m.

ROOM 17, CONVENTION HALL

Blood Volume

1. **J. LaRue Wiley** (*introduced by* H. C. Bazett), **Michael Newton** (*introduced by* H. C. Bazett) **and J. B. Tracy, Jr.** (*introduced by* H. C. Bazett), *Dept. of Physiology, Medical School, University of Pennsylvania*

Relationship in the dog between inferior vena caval pressure and total plasma protein.

2. **C. H. William Ruhe and Paul L. McLain** (*introduced by* C. C. Guthrie), *Dept. of Physiology and Pharmacology, School of Medicine, University of Pittsburgh*

A comparison of heat-coagulable serum solids with serum protein determined by a Kjeldahl method.

3. **J. P. Henry, H. Jacobs** (*by invitation*), **H. Meeham** (*by invitation*) **and A. Karstens** (*by invitation*), *Dept. of Physiology, University of Southern California, and Aero Medical Laby., Air Materiel Command, Wright Field*

The effect of pressure breathing on circulating blood volume. (1) Fluid loss into the tissue spaces.

4. **Thomas H. Allen and Peter D. Orahovats** (*by invitation*), *Dept. of Physiology, College of Physicians and Surgeons, Columbia University*

A method for estimating traces of T-1824 by combination with cellophane.

5. **Robert T. Nieset** (*introduced by* H. S. Mayerson), *School of Medicine, Tulane University, and the Research Division of the Alton Ochsner Medical Foundation*

Blood volume determination by plasma dye dilution and dilution of red cells tagged with P 32.

6. **A. H. Tuttle** (*introduced by* R. R. Overman), *Dept. of Physiology, University of Tennessee College of Medicine*

Blood, plasma and extracellular fluid volumes in experimental low colonic obstruction.

7. **Paul L. McLain and C. H. William Ruhe** (*introduced by* C. C. Guthrie), *Dept. of Physiology and Pharmacology, School of Medicine, University of Pittsburgh*

On estimating the relative volumes of corpuscles and serum in blood.

8. **James C. Moore and O. W. Shadle** (*introduced by* Hampden C. Lawson), *Dept. of Physiology, University of Louisville School of Medicine*

Estimation of the total circulating red cell volume by the use of methemoglobin-tagged cells.

9. **O. W. Shadle and James C. Moore** (*introduced by* Hampden C. Lawson), *Dept. of Physiology, University of Louisville School of Medicine*
   Measurement of red-cell volume loss by the use of methemoglobin-tagged cells.
10. **R. L. Post** (*by invitation*) **and C. R. Spealman,** *Dept. of Physiology, University of Pennsylvania Medical School*
    Variation of total circulating hemoglobin and reticulocyte count with season and following hemorrhage.

PHYSIOLOGY

Papers Read by Title

1. **E. F. Adolph,** *Dept. of Physiology, University of Rochester*
   Cold tolerance and cold immersion in infant rats.
2. **Grant R. Bartlett** (*introduced by* William G. Clark), *Scripps Metabolic Clinic, La Jolla*
   Molecular structure and activity of "vitamin P"-like substances. Inhibition of succinoxidase.
3. **James H. Birnie** (*by invitation*), **W. J. Eversole** (*by invitation*) **and Robert Gaunt,** *Dept. of Zoology, Syracuse University*
   Survival of adrenalectomized-nephrectomized rats treated with desoxycorticosterone.
4. **Stanley Block** (*by invitation*), **Louis Rosenberg** (*by invitation*), **R. H. Broh-Kahn** (*by invitation*) **and I. Arthur Mirsky,** *May Institute for Medical Research of the Jewish Hospital, Cincinnati*
   The source of uropepsin in man.
5. **William J. Bowen** (*introduced by* Heinz Specht), *Laboratory of Physical Biology, National Institute of Health, Bethesda*
   Alkali decomposition of myoglobin.
6. **Daniel A. Brody** (*by invitation*), **J. D. Lawson** (*by invitation*) **and J. P. Quigley,** *Dept. of Physiology, University of Tennessee*
   Production of waves of inhibition in the esophageal-fundic region.
7. **F. Brown** (*by invitation*) **and S. Rodbard,** *Cardiovascular Dept., Research Institute, Michael Reese Hospital, Chicago*
   Pulmonary arterial pressure of the chicken.
8. **L. D. Carlson, A. W. Martin, and V. Gattone** (*by invitation*), *Dept. of Physiology and Biophysics, School of Medicine, University of Washington*
   An apparatus for the measurement of pulmonary function.
9. **T. S. Chang** (*by invitation*) **and Smith Freeman,** *Dept. of Experimental Medicine, Northwestern University and Division of Experimental Medicine, Mayo Foundation*
   Changes in blood level of citric acid and calcium in nephrectomized dogs.
10. **John L. Chapin** (*by invitation*) **and Hermann Rahn,** *Dept. of Physiology, University of Rochester School of Medicine and Dentistry*
    Effect of CO_2 upon the ventilation response of the beaver.
11. **George P. Child** (*by invitation*), **B. S. Hardman** (*by invitation*), **R. A. Woodbury and R. Torpin** (*by invitation*), *Depts. of Pharmacology and Obstetrics and Gynecology, University of Georgia School of Medicine*
    Ephedrine effect on human uterine contractions.
12. **William G. Clark and T. A. Geissman** (*by invitation*), *Scripps Metabolic Clinic, La Jolla, and Dept. of Chemitgry, University of California*
    Molecular structure and activity of "vitamin P"-like substances. Inhibition of oxidation of epinephrine.
13. **J. M. Crismon,** *Dept. of Physiology, Stanford University School of Medicine*
    Errors induced by phosphate in flame photometer analysis of tissue ash for sodium and potassium.
14. **C. G. Drake** (*by invitation*) **and G. W. Stavraky,** *Dept. of Physiology, Faculty of Medicine, University of Western Ontario*
    Effect of convulsant agents on partially isolated neurons of the central nervous system.
15. **Janet Fairfield** (*introduced by* E. F. Adolph), *Dept. of Physiology, University of Rochester*
    Effects of cold on infant rats.
16. **A. M. Freedman,** (*by invitation*) **and H. E. Himwich,** *Medical Division, Army Chemical Center, Md.*
    Effect of size, sex and pregnancy on the lethality of di-isopropyl fluorophosphate (DFP).
17. **M. Friedman and S. O. Byers** (*by invitation*), *The Harold Brunn Institute for Cardiovascular Research, Mt. Zion Hospital, San Francisco*
    Comparison of the renal clearances of allantoin and inulin in man.
18. **M. H. F. Friedman and J. E. Thomas,** *Dept. of Physiology, Jefferson Medical College*
    Preparation and assay of secretin.
19. **Frederick A. Fuhrman,** *Dept. of Physiology, Stanford University School of Medicine*
    Effect of rutin in experimental frostbite.
20. **R. G. Grenell, B. Moore** (*by invitation*), **H. S. Burr** (*by invitation*), **W. Brown** (*by invitation*) **and S. Friedman** (*by invitation*), *Divisions of Neuroanatomy and Psychiatry, Yale University School of Medicine and Fairfield State Hospital, Newtown, Conn.*
    Electrical correlates of psychiatric disturbances.

21. **R. E. Gosselin** (*introduced by* E. E. Adolph), *Dept. of Physiology, University of Rochester*
Acute hypothermia in guinea pigs.

22. **Richard A. Groat,** *Dept. of Anatomy, Bowman Gray School of Medicine of Wake Forest College*
Relationship of volumetric rate of blood flow to arterial diameter.

23. **C. C. Guthrie and Marian E. Lee** (*by invitation*), *Dept. of Physiology and Pharmacology, School of Medicine, University of Pittsburgh*
Quantitative effect of hemorrhage on plasma proteins.

24. **Frederick S. Hammett,** *The Lankenau Hospital Research Institute, Philadelphia*
Possible influence of cosmic energy cycles on growth.

25. **William A. Hiestand and Forst D. Fuller** (*by invitation*), *Laboratory of Animal Physiology, Purdue University, and Dept. of Zoology, DePauw University*
Is heat death due to a myotoxic factor?

26. **Robert Hodes,** *Dept. of Physical Medicine, Graduate School of Medicine, and the Eldridge Reeves Johnson Foundation, University of Pennsylvania*
Muscle action potentials in human poliomyelitis before and after treatment by Billig's method.

27. **S. M. Horvath, R. N. Miller** (*by invitation*) **and B. K. Hutt** (*by invitation*), *Dept. of Physical Medicine, Graduate School of Medicine, University of Pennsylvania*
Heating of human muscle tissue by micro waves.

28. **C. Riley Houck,** *Depts. of Physiology, New York University College of Medicine and the University of Tennessee College of Medicine*
Statistical analysis of filtration rate and renal plasma flow in normal dog and man.

29. **O. H. Janton** (*by invitation*), **H. P. Redondo** (*by invitation*) **and J. C. Scott,** *Dept. of Physiology, The Hahnemann Medical College*
Pulmonary gas exchange following ligation of a pulmonary artery in man.

30. **E. Kaplan and N. R. Joseph** (*introduced by* C. I. Reed), *Dept. of Physiology, College of Medicine and Department of Chemistry, College of Pharmacy, University of Illinois*
Determination of circulation rate in articular structures.

31. **Elizabeth N. King** (*by invitation*), **M. H. F. Friedman and I. J. Pincus** (*by invitation*), *Dept. of Physiology, Jefferson Medical College, Philadelphia*
Tryptic activity of pancreatic juice after dilution with gastric juice.

32. **Bruno Kisch,** *Marine Biological Laboratory, Woods Hole*
The electrogram of the fish heart.

33. **Alfred Leimdorfer,** *Dept. of Pharmacology, University of Illinois College of Medicine*
Electroencephalographic analysis of action of amidone, morphine and strychnine on the central nervous system.

34. **D. H. LeMessurier** (*introduced by* C. N. Woolsey), *Dept. of Physiology, Johns Hopkins University School of Medicine*
Auditory and visual areas of the cerebral cortex of the rat.

35. **Richard W. Lippman** (*introduced by* T. Addis), *Dept. of Medicine, Stanford University School of Medicine*
Effects of protein and fluid consumption upon plasma volume and circulating protein in the rat.

36. **David I. Macht,** *Division of Pharmacology, Laboratories of Sinai Hospital, Baltimore*
Thromboplastic effects of sulfa and penicillin combinations.

37. **Martin B. Macht and Elizabeth L. Pillion** (*introduced by* H. S. Belding), *The Quartermaster Corps, Climatic Research Laboratory, Lawrence, Mass.*
Changes in skin temperature and blood flow of hand following ingestion of certain amino acids.

38. **N. S. R. Maluf,** *Dept. of Pharmacology, University of Louisville School of Medicine and the Louisville General Hospital*
Urea clearance by perfusion of the entire intact small intestine in man.

39. **John Marshall** (*by invitation*) **and G. E. Wakerlin,** *Dept. of Physiology, University of Illinois College of Medicine*
Purification of renin.

40. **E. M. Martin** (*by invitation*), **M. J. Martin** (*by invitation*) **J. P. Henry, J. L. Gamble, Jr.** (*by invitation*) **and R. S. Shaw** (*by invitation*), *Aero Medical Laboratory, Air Materiel Command, Wright Field*
The effects of various pressure application devices (anti-g suits) upon arterial and venous pressures.

41. **A. T. Milhorat,** *Depts. of Psychiatry and Medicine, Cornell University Medical College, The Russell Sage Institute of Pathology and the New York Hospital*
Effect of delta- and gamma-tocopherol on creatinuria in progressive muscular dystrophy.

42. **Dorothy Nelson** (*introduced by* J. S. Gray), *Dept. of Physiology, Northwestern University Medical School*
The sodium chloride tolerance of female mice.

43. **John F. Perkins, Jr. and M. C. Li** (*introduced*

by E. M. Landis), *Dept. of Physiology, Harvard Medical School*

The effects of changing the temperature of the normal and denervated nictitating membrane.

44. **J. J. Reinhard, Jr.** (*by invitation*) **O. Glasser** (*by invitation*) **and Irvine H. Page,** *Research Division of the Cleveland Clinic Foundation*

Effects of hemorrhagic shock on hepatectomized and bilaterally nephrectomized-hepatectomized dogs.

45. **Boris B. Rubenstein,** *Dept. of Metabolism-Endocrinology Research, Michael Reese Hospital*

Does progesterone or testosterone alter the human vaginal smear?

46. **Gerald R. Seaman** (*introduced by* C. G. Wilber) *Biological Laboratory, Fordham University*

Relation between growth and synthesis of lipids in colpidium.

47. **Herbert Shapiro,** *Laboratory of Physical Biology, National Institute of Health, Bethesda*

The retarding action of vitamin C in physiological concentrations on the rate of cell division.

48. **Walter B. Shelley and Frank N. Melton** (*by invitation*), *Dept. of Dermatology, University of Pennsylvania School of Medicine*

The permeability of normal human skin to histamine.

49. **Ephraim Shorr, B. W. Zweifach and S. Baez** (*by invitation*), *Dept. of Medicine, Cornell University Medical College and the New York Hospital*

Hepato-renal factors in circulatory homeostasis.

50. **Franklin F. Snyder,** *Harvard Medical School*

Oxygenation of the cord blood of breathing fetuses during a prolonged period.

51. **F. J. Sichel and Cheryl Parkhurst** (*by invitation*), *University of Vermont College of Medicine*

The leakage of potassium from injured muscle fibres.

52. **Verne W. Swigert** (*by invitation*), **Hamilton R. Fishback** (*by invitation*), **Lillian E. Cisler** (*by invitation*), **and Frederic T. Jung,** *Northwestern University Medical School*

Silicone implantations in the rat.

53. **Julian M. Tobias,** *Dept. of Physiology and Toxicity Laboratory, University of Chicago*

Sodium and potassium in insects: larvae, pupae and adults.

54. **Clara Torda and Harold G. Wolff,** *New York Hospital and Depts. of Medicine (Neurology) and Psychiatry, Cornell University Medical College*

Effect of acetylcholine, caffeine and alkaloids on activity of muscle adenosinetriphosphatase.

55. **Frank Visscher,** *Research Laboratories, Upjohn Co., Kalamazoo*

Fractionation studies of enterogastrone activity in pyloric ligation rats.

56. **G. van Wagenen,** *Dept. of Obstetrics and Gynecology, Yale University School of Medicine*

Vitamin A deficiency in pregnancy.

57. **Virginia Weimar** (*by invitation*) **and Rosalind Wulzen,** *Dept. of Zoology, Oregon State College*

Leucocytosis of guinea pigs deficient in the anti-stiffness factor.

58. **Arne N. Wick and Francis Pauls** (*introduced by* Eaton N. MacKay), *Scripps Metabolic Clinic, La Jolla*

Preparation of anti-ulcer substance from bovine urine.

59. **Charles G. Wilbur,** *The Biological Laboratory, Fordham University*

Glucose metabolism in marine invertebrates.

60. **D. L. Wilson** (*by invitation*) **and J. F. Manery,** *Dept. of Biochemistry, University of Toronto*

Sodium, potassium and chloride in leucocytes.

61. **Claude V. Winder, Mona M. Anderson** (*by invitation*) **and Hervey C. Parke** (*by invitation*), *Research Laboratories, Parke, Davis & Co, Detroit*

Comparative properties of six phenethylamines.

62. **Verner J. Wulff** (*by invitation*) **and Theodore L. Jahn,** *Dept. of Zoology and Physiology, University of Illinois, and Depts. of Physiology and Zoology, State University of Iowa*

Relation of retinal and optic nerve response to intensity of illumination of the grasshopper eye.

63. **B. W. Zweifach, S. Baez** (*by invitation*), **R. F. Furchgott** (*by invitation*) **and Ephraim Shorr,** *Dept. of Medicine, Cornell University Medical College and the New York Hospital*

Hepato-renal factors in circulatory homeostasis.

AMERICAN SOCIETY OF BIOLOGICAL CHEMISTS, INC.

THIRTY-NINTH ANNUAL MEETING

BIOCHEMISTRY A

Tuesday, March 16, 9:00 a.m.

ROOM C, CONVENTION HALL

Acetate Metabolism

1. **Albert L. Lehninger and Eugene P. Kennedy** (*by invitation*), *Depts. of Surgery and Biochemistry, University of Chicago*
   The fatty acid oxidase complex of rat liver and intracellular structures.
2. **Nathan O. Kaplan** (*by invitation*) **and Fritz Lipmann,** *Biochemical Research Lab., Massachusetts General Hospital, and the Dept. of Biological Chemistry, Harvard University Medical School, Boston*
   Reactions between acetate, acetyl phosphate and the adenylic acid system in tissue and bacterial extracts.
3. **Harold Strecker** (*by invitation*), **L. O. Krampitz, and Harland G. Wood,** *Dept. of Biochemistry, Western Reserve University School of Medicine, Cleveland*
   The role of acetylphosphate in the phosphoclastic and dismutation reactions of pyruvate.
4. **David Novelli** (*by invitation*) **and Fritz Lipmann,** *Biochemical Research Lab., Massachusetts General Hospital, and the Dept. of Biological Chemistry, Harvard University Medical School, Boston*
   Respiratory metabolism in yeast and coenzyme A levels. Effect of phenyl panthenone on coenzyme A synthesis.
5. **Michael S. Weiss, Helen Voripaieff, and David Nachmansohn** (*introduced by* H. T. Clarke), *Dept. of Neurology, College of Physicians and Surgeons, Columbia University, New York*
   Inhibitors of choline acetylase.
6. **Morris Soodak** (*by invitation*) **and Fritz Lipmann,** *Biochemical Research Lab., Massachusetts General Hospital, and the Depts. of Biological Chemistry and Medicine, Harvard University Medical School, Boston*
   An enzymatic micromethod for determination of acetic acid.
7. **Simon Black** (*introduced by* E. S. Guzman Barron), *Chemical Div., Dept. of Medicine, University of Chicago*
   A microanalytical method for acetic and other volatile acids.
8. **Konrad Bloch and W. Kramer** (*by invitation*), *Dept. of Biochemistry and Inst. of Radiobiology and Biophysics, University of Chicago*
   Synthesis of fatty acids in rat liver slices.
9. **H. S. Anker** (*introduced by* E. A. Evans, Jr.), *Dept. of Biochemistry, University of Chicago*
   On the fate of labeled pyruvic acid in the intact animal.
10. **R. Gordon Gould** (*by invitation*), **I. M. Rosenberg** (*by invitation*), **Marott Sinex** (*by invitation*), **and A. Baird Hastings,** *Dept. of Biological Chemistry, Harvard University Medical School, Boston*
    Rate of $C^{14}O_2$ excretion following intraperitoneal administration of isotopic bicarbonate and acetate.
11. **W. D. Armstrong, Jack Schubert** (*by invitation*), **and Arthur Lindenbaum** (*by invitation*), *Dept. of Physiological Chemistry, University of Minnesota Medical School, Minneapolis*
    Tissue incorporation and excretion of radioactive carbon administered as carbonate.

BIOCHEMISTRY B

Tuesday, March 16, 9:00 a.m.

ROOM D, CONVENTION HALL

Amino Acid Determinations

1. **Stanford Moore** (*by invitation*) **and William H. Stein,** *The Rockefeller Institute for Medical Research, New York*
   Chromatography of amino acids. Colorimetric ninhydrin method for analysis of the effluent.
2. **William H. Stein and Stanford Moore** (*by invitation*), *The Rockefeller Institute for Medical Research, New York*
   Chromatography of amino acids. Solvent systems for the fractionation of protein hydrolysates.
3. **Albert S. Keston** (*by invitation*), **Sidney Udenfriend** (*by invitation*), **and R. Keith Cannan,** *Dept. of Chemistry, New York University College of Medicine*
   Application of the isotopic derivative method of analysis to protein hydrolysates.
4. **Milton Levy and Evelyn Slobodiansky** (*by invitation*), *Dept. of Chemistry, New York University College of Medicine*
   The arrangement of amino acids in silk; an application of the isotopic derivative technique.

5. **Albert S. Keston** (*by invitation*), **Sidney Udenfriend** (*by invitation*), **and Milton Levy,** *Dept. of Chemistry, New York University College of Medicine, New York*
   Quantitative analysis of protein hydrolysates on paper chromatograms by means of the isotopic derivative method.
6. **N. F. Young and F. Homburger** (*introduced by* T. F. Gallagher), *The Sloan-Kettering Inst. for Cancer Research, Research Div. Memorial Cancer Center, New York*
   The application of paper chromatography to the study of aminoaciduria in patients with liver disease.
7. **Stanley R. Ames and Hugh A. Risley** (*introduced by* P. L. Harris), *Labs. of Distillation Products, Inc., Rochester*
   Aminoaciduria in progressive muscular dystrophy.
8. **J. Logan Irvin and Elinor Moore Irvin** (*by invitation*), *Dept. of Physiological Chemistry, Johns Hopkins University School of Medicine, Baltimore*
   The determination of various amines by proton-exchanges with ethyl eosin in non-aqueous solvents.
9. **M. X. Sullivan,** *Chemo-Medical Research Inst., Georgetown University, Washington, D. C.*
   A comparison of reactions of cysteine and Beta-beta dimethyl cysteine.
10. **Dean Burk, Silvio Fiala** (*by invitation*), **John Hearon** (*by invitation*), **Marie Hesselbach** (*by invitation*), **Milton Levy** (*by invitation*), **and Arthur L. Schade,** *National Cancer Institute, National Institute of Health, Bethesda, Md., and Overly Biochemical Research Foundation, New York*
    Reversible oxygenation, irreversible oxidation, and decarboxylation of cobalt-amino acid complexes with oxygen gas.
11. **Martin E. Hanke,** *Dept. of Biochemistry, University of Chicago*
    Conditions necessary for complete decarboxylation of pure L-lysine and L-tyrosine by amino decarboxylases.

BIOCHEMISTRY C

Tuesday, March 16, 9:00 a.m.

ROOM B, CONVENTION HALL

Proteins

1. **Sidney Roberts** (*by invitation*) **and Abraham White,** *Dept. of Physiological Chemistry, Yale University School of Medicine, New Haven*
   Biochemical characterization of lymphoid tissue proteins.
2. **Erland C. Gjessing** (*introduced by* Alfred Chanutin), *Biochemical Lab., University of Virginia, Richmond*
   Fractionation of lymphoid tissues.
3. **Adolph Abrams** (*by invitation*) **and Philip P. Cohen,** *Dept. of Physiological Chemistry, University of Wisconsin Medical School, Madison*
   Fractionation studies on the cytoplasmic proteins of human lymphoid tissue.
4. **Jordi Folch and L. L. Uzman** (*by invitation*), *McLean Hospital, Waverley, Mass., and Harvard University Medical School, Boston*
   Brain proteins: isolation of a birefringent liponucleoprotein.
5. **J. Murray Luck and A. Clark Griffin** (*by invitation*), *Dept. of Chemistry, Stanford University, Calif.*
   Fractionation of liver proteins.
6. **Frank W. Putnam, Lloyd M. Kozloff** (*by invitation*), **and E. A. Evans, Jr.,** *Dept. of Biochemistry, University of Chicago*
   Purification and properties of *E. coli* bacteriophage T_6.
7. **D. W. Woolley,** *Labs. of The Rockefeller Institute for Medical Research, New York*
   Some crystalline peptides isolated from enzymic digests of insulin and their relationship to strepogenin.
8. **Frank A. Csonka,** *Bureau of Human Nutrition and Home Economics, Agricultural Research Administration, U. S. Dept. of Agriculture, Washington*
   Dietary influence on the amino acid composition of proteins.
9. **T. L. McMeekin, B. D. Polis** (*by invitation*), **E. S. Della Monica** (*by invitation*), **and J. H. Custer** (*by invitation*), *Eastern Regional Research Lab., Philadelphia*
   A crystalline compound of β-lactoglobulin with dodecyl sulfate.
10. **Stephan Ludewig and Alfred Chanutin,** *Biochemical Lab., University of Virginia, Charlottesville*
    Studies on the agglutination of human and rat red cells by castor bean extracts.

BIOCHEMISTRY D

Tuesday, March 16, 9:00 a.m.,

ROOM S, CONVENTION HALL

Lipids

1. **Walter Marx and Mortimer Lipsett** (*introduced by* Harry J. Deul, Jr.), *Dept. of Biochemistry, University of Southern California Medical School, Los Angeles*

Enzymatic destruction of cholesterol by rat liver extract *in vitro*.

2. **R. T. Holman** (*introduced by* P. B. Pearson), *Medicinska Nobelinst., Stockholm, and Dept. of Biochemistry and Nutrition, A and M College of Texas, College Station*
Coupled oxidations in enzymatically oxidized linoleic acid.
3. **Camillo Artom and Marjorie A. Swanson** (*by invitation*), *Dept. of Biochemistry, Bowman Gray School of Medicine, Wake Forest College, Winston-Salem*
On the absorption of phospholipides.
4. **Camillo Artom and W. E. Cornatzer** (*by invitation*), *Dept. of Biochemistry, Bowman Gray School of Medicine, Wake Forest College, Winston-Salem*
The action of ethanolamine, monomethyl- and dimethyl-ethanolamine on lipide phosphorylation.
5. **Warren M. Sperry and Florence C. Brand** (*by invitation*), *Dept. of Biochemistry, New York State Psychiatric Institute, New York*
The determination of choline in brain lipids.
6. **Sie Hsien-gieh** (*by invitation*), **C. E. Faust** (*by tion*), **and Harold H. Williams,** *Dept. of Biochemistry, Cornell University, Ithaca*
Studies on cerebrosides.
7. **Alfred E. Koehler, Elsie Hill** (*by invitation*), **and Frank Fearney** (*by invitation*), *Santa Barbara Cottage Hospital and The Sansum Clinic Research Foundation, Santa Barbara*
Studies on the micro molecular distillation of blood lipids.
8. **J. C. Forbes and William B. Porter** (*by invitation*), *Depts. of Biochemistry and Medicine, Medical College of Virginia, Richmond*
Fractionation of serum cholesterol.

JOINT SESSION OF THE FEDERATION

Tuesday, March 16, 1:30 p.m.

Ballroom, Convention Hall

Program on page 314

BIOCHEMISTRY BUSINESS MEETING

Tuesday, March 16, 4:15 p.m.

Room C

BIOCHEMISTRY A

Wednesday, March 17, 9:00 a.m.

Room C, Convention Hall

Phosphorus Compounds

1. **Otto Meyerhof and Peter Oesper** (*by invitation*), *Dept. of Physiological Chemistry, School of Medicine, University of Pennsylvania, Philadelphia*
The free energy of phosphorylation.
2. **Ernst Vischer** (*by invitation*) **and Erwin Chargaff,** *Dept. of Biochemistry, College of Physicians and Surgeons, Columbia University, New York*
Studies on the composition of nucleic acids.
3. **Gerhard Schmidt, Ricardo Cubiles** (*by invitation*), **and S. J. Thannhauser,** *Boston Dispensary, Tufts College Medical School*
Ribopolynucleotide fractions formed during the enzymatic hydrolysis of yeast ribonucleic acid
4. **Zacharias Dische,** *Dept. of Biochemistry, College of Physicians and Surgeons, Columbia University, New York*
The breakdown of free and combined adenosine-5-phosphate (AdPh) in intact human erythrocytes.
5. **E. Racker** (*introduced by* S. Ochoa), *Dept. of Bacteriology, New York University College of Medicine*
Enzymatic formation and breakdown of pentose phosphate.
6. **W. Wayne Kielley** (*introduced by* Otto Meyerhof), *Dept. of Physiological Chemistry, School of Medicine, University of Pennsylvania, Philadelphia*
A new adenosinetriphosphatase of muscle.
7. **Arthur Kornberg and Olov Lindberg** (*introduced by* Carl F. Cori), *Dept. of Biological Chemistry, Washington University School of Medicine, St. Louis*
DPN pyrophosphatase.
8. **Aser Rothstein and Rebecca Meier** (*introduced by* Harvey B. Haag), *University of Rochester School of Medicine and Dentistry, N. Y.*
Adenylpyrophosphatases and other phosphatases in the cell surface of living yeast. (Pharmacol.)
9. **H. G. Albaum, M. Ogur** (*by invitation*), **H. Mendelow** (*by invitation*), **and A. Hirshfeld** (*by invitation*), *Depts. of Biology and Chemistry of Brooklyn College*
Adenine-pentose-pyrophosphate from mung beans.
10. **Oscar Bodansky and Norma Strachman** (*by invitation*), *Dept. of Pharmacology, Cornell University Medical College, New York*
The inhibitory effects of α-amino acids on phosphatase activity.
11. **Eunice V. Flock and Jesse L. Bollman,** *Mayo Foundation, Rochester, Minn.*
Alkaline phosphatase activity of intestinal lymph of the rat.

BIOCHEMISTRY B

Wednesday, March 17, 9:00 a.m.

Room D, Convention Hall

Amino Acid Metabolism

1. **H. R. Crookshank** (*by invitation*) **and Clarence P. Berg,** *Biochemistry Dept., Medical College of Alabama, Birmingham, and the Biochemical Lab., State University of Iowa, Iowa City*
   Changes in the composition of the blood of rats fed L-histidine.
2. **Ella H. Fishberg,** *Biochemical Lab. of Beth Israel Hospital, New York*
   The quinoid stage of tyrosine metabolism.
3. **W. Knowlton Hall** (*by invitation*), **V. P. Sydenstricker and Katrine Rawls** (*by invitation*), *Depts. of Medicine and of Biochemistry, University of Georgia School of Medicine, Augusta*
   Factors affecting the excretion of metabolites of phenylalanine and tyrosine in alkaptanuria. (Nutrition)
4. **Halvor N. Christensen,** *The Children's Hospital, and the Dept. of Biological Chemistry, Harvard University Medical School, Boston*
   The distribution of amino acids between fetal and maternal extracellular fluids and cells.
5. **Philip Handler, Henry Kamin** (*by invitation*), **and Jerome S. Harris** (*by invitation*), *Depts. of Biochemistry and Pediatrics, Duke University School of Medicine, Durham*
   The metabolism of parenterally administered amino acids. I. Glycine.
6. **David Shemin, Irving M. London** (*by invitation*), **and D. Rittenberg,** *Depts. of Biochemistry and Medicine, College of Physicians and Surgeons, Columbia University, New York*
   The *in vitro* synthesis of heme from glycine by the nucleated red blood cell.
7. **Irving M. London** (*by invitation*), **Randolph West, David Shemin, and D. Rittenberg,** *Depts. of Medicine and Biochemistry, College of Physicians and Surgeons, Columbia University, New York, and the Presbyterian Hospital in the City of New York*
   On the origin of stercobilin in humans.
8. **Louis D. Greenberg and James F. Rinehart** (*by invitation*), *Divs. of Pathology and Pharmacology, University of California Medical School, San Francisco*
   Xanthurenic acid excretion in pyridoxine deficient rhesus monkeys.
9. **Jack Schultz and George T. Rudkin** (*introduced by* Gerit Toennies), *Lankenau Hospital Research Inst., and Inst. for Cancer Research, Philadelphia*
   Absence of a sparing action of tryptophane on nicotinamide requirements of the fly, *Drosophila melanogaster*.
10. **Robert MacVicar** (*by invitation*) **and R. H. Burris,** *Dept. of Biochemistry, University of Wisconsin, Madison*
    Studies on the nitrogen metabolism of tomato using N^{15}-labeled ammonium sulfate.
11. **James M. Orten,** *Dept. of Physiological Chemistry, Wayne University College of Medicine, Detroit*
    Further studies on the effect of cysteine and histidine on the production of cobalt polycythemia.

BIOCHEMISTRY C

Wednesday, March 17, 9:00 a.m.

Room B, Convention Hall

Liver Protein Regeneration, Nitrogen Balance

1. **D. Rittenberg, Edith E. Sproul** (*by invitation*), **and David Shemin,** *Dept. of Biochemistry and Pathology, College of Physicians and Surgeons, Columbia University, New York*
   Rate of protein formation in the livers of partially hepatectomized rats.
2. **Harry M. Vars and Charles E. Friedgood** (*by invitation*), *Harrison Dept. of Surgical Research, School of Medicine, University of Pennsylvania, Philadelphia*
   The influence of fat in the diet upon nitrogen metabolism and liver protein regeneration.
3. **Leon L. Miller,** *Dept. of Biochemistry, Jefferson Medical College, Philadelphia*
   Loss and restoration of rat liver enzymes (activity) related to dietary changes in liver protein.
4. **Sam Seifter, David M. Harkness** (*by invitation*), **B. Feldman** (*by invitation*), **Leonard Rubin** (*by invitation*), **and Edward Muntwyler,** *Dept. of Biochemistry, Long Island College of Medicine, Brooklyn*
   Concentration changes of certain vitamins and enzymes in livers of rats on protein-free diet.
5. **Carl Alper** (*by invitation*), **Shirley DeBiase** (*by invitation*), **and Bacon F. Chow,** *Div. of Protein Chemistry, The Squibb Inst. for Medical Research, New Brunswick*
   The rate of disappearance of human albumin in normal and protein depleted dogs.
6. **W. C. Hess,** *Georgetown Medical School, Washington*
   The rates of absorption of L- and DL-methionine.
7. **James D. Solomon and Stanley W. Hier** (*introduced by* Olaf Bergeim), *Dept. of Biological Chemistry, University of Illinois College of Medicine, and the Research Labs., The Wilson Labs., Chicago*
   Influence of feeding amino acids on the spinal fluid level of free amino acids.

8. **Robert H. Silber, E. E. Howe, and Curt C. Porter** (*introduced by* Edgar G. Miller), *Merck Inst. for Therapeutic Research and the Merck Research Labs., Rahway, N. J.*
The urinary excretion of amino acids and peptides after intravenous infusion to dogs.
9. **H. E. Sauberlich** (*by invitation*) **and C. A. Baumann,** *Dept. of Biochemistry, College of Agriculture, University of Wisconsin, Madison*
Amino acid excretion as influenced by dietary proteins of different biological value.
10. **George S. Samuelsen** (*by invitation*), **Grace E. Griffin** (*by invitation*), **Lois E. Griffith** (*by invitation*), **Sam Seifter and Edward Muntwyler,** *Dept. of Biochemistry, Long Island College of Medicine, Brooklyn*
Blood and "tissue" protein changes in dogs on protein-deficient diets with and without supplementation.

BIOCHEMISTRY D

Wednesday, March 17, 9:00 a.m.

ROOM S, CONVENTION HALL

Steroids

1. **H. Hirschmann, Frieda B. Hirschmann** (*by invitation*), **and Margaret A. Daus** (*by invitation*), *Dept. of Medicine, Western Reserve University and the Lakeside Hospital, Cleveland*
Stereochemistry of allopregnanetriol-3,16,20 of mare's urine.
2. **William J. Haines, Mari P. Goodwin, George Pish, and Foil A. Miller** (*introduced by* Marvin H. Kuizenga), *Research Labs., The Upjohn Co., Kalamazoo, Mich., and Div. of Physical Chemistry, University of Illinois, Urbana*
An infra-red and x-ray study of polymorphism in 5-pregnen-3(β)ol-20-one.
3. **W. H. Pearlman,** *Dept. of Biochemistry, Jefferson Medical College of Philadelphia*
The isolation of pregnanol-3(α)-one-20, comp'd Y ($C_{21}H_{36}O_2$) and comp'd Z ($C_{19}H_{32}O_2$) from the bile of pregnant cows.
4. **Seymour Lieberman** (*by invitation*), **David K. Fukushima** (*by invitation*), **and Konrad Dobriner,** *Sloan-Kettering Inst. for Cancer Research*
Adrenal cortical metabolites in human urine.
5. **Benjamin F. Stimmel,** *Rees-Stealy Medical Research Fund, San Diego*
The effect of zinc-hydrochloric acid hydrolysis on the estrogens in human urine.
6. **Harold J. Nicholas** (*by invitation*), **Sidney A. Thayer, and Edward A. Doisy,** *Lab. of Biological Chemistry, St. Louis University School of Medicine*
Estrogenic activity of doisynolic acid and related compounds.
7. **Ernest J. Umberger and Jack M. Curtis** (*introduced by* Arnold J. Lehman), *Div. of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington,* D. C.
The estimation of alpha and beta estradiols and of estrone and equilin in binary mixtures. (Pharmacol.)
8. **Blaine H. Levedahl** (*by invitation*) **and Leo T. Samuels,** *Dept. of Biochemistry, University of Utah School of Medicine, Salt Lake City*
The effect of testosterone and methyl testosterone on guanidoacetic acid in blood and urine.
9. **John J. Schneider** (*by invitation*) **and Harold L. Mason,** *Div. of Biochemistry, Mayo Clinic, Rochester*
Substances isolated following the incubation of dehydroisoandrosterone, androsterone and etiocholanolone with liver tissue.
10. **Max L. Sweat** (*by invitation*) **and L. T. Samuels,** *Dept. of Biochemistry, University of Utah School of Medicine, Salt Lake City*
An enzyme requiring DPN involved in the metabolism of testosterone by liver tissue.
11. **Louis Levin and Joseph W. Jailer** (*by invitation*), *Depts. of Anatomy and Obstetrics and Gynecology, College of Physicians and Surgeons, Columbia University, New York*
The effect of gonadotrophic stimulation on the cholesterol content of the immature rat ovary.
12. **G. A. Wills, Sybil Rampton** (*by invitation*) **and L. I. Pugsley,** *Food and Drugs Div., Dept. National Health and Welfare, Ottawa, Canada*
Variables affecting the assay of testosterone propionate using male castrate rats.
13. **Lewis L. Engel, Helen R. Patterson and Hildegard Wilson** (*introduced by* A. Baird Hastings), *Medical Labs. of the Collis P. Huntington Memorial Hospital of Harvard University, at the Massachusetts General Hospital, Boston, and the Dept. of Biological Chemistry, Harvard University Medical School, Boston*
The quantitative estimation of steroid alcohols.

BIOCHEMISTRY

Wednesday, March 17, 1:45 p.m.

ROOM C, CONVENTION HALL

Symposium on Methods for the Determination of the Purity of Substances of Biochemical Interest

VINCENT DU VIGNEAUD, *Chairman*

1. **Dr. George H. Cassidy,** *of Yale University.*
   Chromatography.
2. **R. Bowling Barnes,** *American Cyanamid Company, Stamford, Conn.*
   Spectrophotometry.
3. **Lyman C. Craig,** *The Rockefeller Inst. for Medical Research, New York*
   Counter-current distribution.
4. **J. W. Williams,** *University of Wisconsin, Madison*
   Electrophoresis and ultracentrifugation.
5. **R. M. Herriott,** *The Rockefeller Inst. for Medical Research, Princeton*
   Solubility analysis.
6. **James Hillier,** *RCA Labs., Princeton*
   Electron microscopy.

Federation "Mixer"

Wednesday, March 17, 9:00 p.m.

ARENA, CONVENTION HALL

BIOCHEMISTRY A

Thursday, March 18, 9:00 a.m.

ROOM C, CONVENTION HALL

Carbohydrate Metabolism

1. **Earl W. Sutherland and Christian deDuve** (*introduced by* Carl F. Cori), *Dept. of Biochemistry, Washington University School of Medicine, St. Louis*
   A glycogenolytic factor from pancreas.
2. **George L. Collins, Jr.** (*by invitation*), **Edward M. Bridge, and Barbara Maitland** (*by invitation*), *Statler Research Labs. of the Children's Hospital, Buffalo, and Dept. of Pediatrics of the University of Buffalo*
   The action of insulin on glycogen reserves.
3. **Michael Somogyi,** *Lab. of the Jewish Hospital of St. Louis*
   A paradoxical effect of insulin on glucose assimilation.
4. **C. A. Villee, F. M. Sinex, and A. K. Solomon** (*introduced by* A. Baird Hastings), *Dept. of Biological Chemistry and the Biophysical Lab., Harvard University Medical School, Boston*
   In vitro utilization of glucose by rat diaphragm muscle.
5. **Sigmund Schwimmer,** *Enzyme Research Lab., Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Dept. of Agriculture, Albany, Calif.*
   Purification of alpha-amylase from barley malt extracts.
6. **Victor A. Najjar,** *Dept. of Biological Chemistry, Washington University School of Medicine, St. Louis*
   The purification and crystallization of phosphoglucomutase.
7. **Edwin G. Krebs** (*by invitation*), **and Victor A. Najjar,** *Dept. of Biological Chemistry, Washington University School of Medicine, St. Louis*
   Immunochemical studies on purified d-glyceraldehyde-3-phosphate dehydrogenase from yeast and rabbit muscle.
8. **Sidney F. Velick,** *Dept. of Biological Chemistry, Washington University School of Medicine, St. Louis*
   An electrophoretic analysis of the interaction of aldolase and glyceraldehyde-phosphate dehydrogenase with phosphate ions.
9. **Karl Meyer and Charles Ragan** (*by invitation*), *Depts. of Ophthalmology and Medicine, School of Medicine, Columbia University, New York*
   Inhibition of hyaluronidase by hydroquinones and quinones.
10. **William H. Fishman, Kurt I. Altman** (*by invitation*), **and Belle Springer** (*by invitation*), *Depts. of Surgery and Biochemistry, University of Chicago*
    Blood plasma anti-glucuronidase activity.

BIOCHEMISTRY B

Thursday, March 18, 9:00 a.m.

ROOM D, CONVENTION HALL

Amino Acid Metabolism (continued)

1. **Theodore Winnick, Felix Friedberg** (*by invitation*), **and David M. Greenberg,** *Div. of Biochemistry, University of California Medical School, Berkeley*
   Incorporation of C^{14} labeled glycine into the protein of tissue homogenates.
2. **Henry Borsook, Clara L. Deasy** (*by invitation*), **Jacob W. Dubnoff, C. T. O. Fong** (*by invitation*), **William D. Fraser** (*by invitation*), **A. J. Haagen-Smit** (*by invitation*), **Geoffrey Keighley** (*by invitation*), **Peter H. Lowry** (*by invitation*), *William G. Kerckhoff Labs. of the Biological Sciences, California Inst. of Technology, Pasadena*
   Protein and peptide turnover with respect to lysine in guinea pig liver homogenate.
3. **Philip P. Cohen and Santiago Grisolia** (*by invitation*), *Dept. of Physiological Chemistry, University of Wisconsin Medical School, Madison*
   The mechanism of enzymatic synthesis of citrulline from ornithine.

4. **Ernest Borek and Heinrich Waelsch,** *Depts. of Biochemistry, New York State Psychiatric Inst. and Columbia University, New York*
The role of bicarbonate in glutamic acid metabolism.

5. **Heinrich Waelsch and Phyllis Owades** (*by invitation*), *Depts. of Biochemistry, New York State Psychiatric Inst. and Columbia University, New York*
The inhibition of glutaminase by glutamic acid.

6. **Alton Meister** (*by invitation*) **and Jesse P. Greenstein,** *National Cancer Inst., National Inst. of Health, Bethesda, Md.*
Enzymatic hydrolysis of acetopyruvic acid.

7. **Vincent E. Price** (*by invitation*) **and Jesse P. Greenstein,** *National Cancer Inst., National Inst. of Health, Bethesda, Md.*
N-acylated and N-methylated glycyldehydroalanine and related compounds.

8. **David B. Sprinson** (*by invitation*) **and D. Rittenberg,** *Dept. of Biochemistry, College of Physicians and Surgeons, Columbia University, New York*
The metabolism of the α, β and γ hydrogen atoms of L-leucine.

9. **Aaron Bunsen Lerner** (*by invitation*), **Thomas B. Fitzpatrick** (*by invitation*), **Evan Calkins** (*by invitation*), **and William H. Summerson,** *Biochemistry Section, Medical Div., Army Chemical Center, Md.*
Enzymatic oxidation of tyrosine and dihydroxyphenylalanine by melanoma extracts.

BIOCHEMISTRY C

Thursday, March 18, 9:00 a.m.

ROOM B, CONVENTION HALL

Pteroylglutamic Acid

1. **Arnold D. Welch, Robert W. Heinle** (*by invitation*), **Jack A. Pritchard** (*by invitation*), **and Herbert Salis,** (*by invitation*), *Depts. of Pharmacology and Medicine, Western Reserve University School of Medicine, Cleveland*
Influence of pteroylglutamic acid on the synthesis and action of the antipernicious anemia factor. (Nutrition)

2. **R. C. Grubbs** (*by invitation*), **B. C. Houghton** (*by invitation*), **Julia Trossbach** (*by invitation*), **and F. A. Hitchcock,** *The Lab. of Avialon Physiology, Ohio State University, Columbus*
The effects of folic acid on respiratory metabolism. (Nutrition)

3. **Paul L. Day, Dorothy S. Gaines** (*by invitation*), **Marion McKee** (*by invitation*), **Phyllis Scroggin** (*by invitation*), **and Raymond Houchins** (*by invitation*), *Dept. of Biochemistry, School of Medicine, University of Arkansas, Little Rock*
Pteroylglutamic acid balance studies on monkeys. (Nutrition)

4. **Conrado F. Asenjo,** *Dept. of Chemistry and Nutrition, School of Tropical Medicine, San Juan, Puerto Rico*
Folic acid requirement of the rat and some characteristic lesions observed in the deficient animals.

5. **Charles W. Mushett** (*by invitation*) **and Gladys A. Emerson,** *Merck Inst. for Therapeutic Research, Rahway, N. J.*
Response of leukopenia and granulocytopenia in sulfathiazole-fed rats to pteroylglutamic acid and to parenteral liver extracts. (Nutrition)

6 **Gladys A. Emerson and Charles W. Mushett** (*by invitation*), *Merck Inst. for Therapeutic Research, Rahway, N. J.*
Influence of liver extracts on a sulfathiazole induced dietary deficiency in rats. (Nutrition)

7. **B. S. Schweigert** (*introduced by* P. B. Pearson), *Dept. of Biochemistry and Nutrition, A and M College of Texas, College Station*
Studies on folic acid conjugase in blood.

8. **M. E. Swendseid** (*by invitation*), **E. L. Wittle** (*by invitation*), **G. W. Moersch** (*by invitation*), **O. D. Bird, and R. A. Brown,** *The Research Labs., Parke, Davis and Co., Detroit*
Studies in the rat of inhibitors of pteroylglutamic acid structurally related to this vitamin. (Nutrition)

9. **E. L. R. Stokstad, M. Regan** (*by invitation*), **A. L. Franklin** (*by invitation*), **and T. H. Jukes,** *Lederle Labs. Div., American Cyanamid Co., Pearl River, N. Y.*
Interrelationships between pteroylglutamic acid, adenine thymine, and antagonists of pteroylglutamic acid.

10. **George H. Hitchings, Gertrude B. Elion** (*by invitation*), **and Henry VanderWerff** (*by invitation*), *The Wellcome Research Labs., Tuckahoe, N. Y.*
a-Aminopurine as a purine antagonist.

11. **John R. Totter and Edith S. Sims** (*by invitation*), *Dept. of Biochemistry, School of Medicine, University of Arkansas, Little Rock*
The influence of KCN and pteroylglutamic acid on growth and porphyrin production of *Corynebacterium hoffmannii*.

BIOCHEMISTRY D

Thursday, March 18, 9:00 a.m.

Room S, Convention Hall

Sulfur Compounds

1. **J. P. Saunders and W. A. Himwich** (*introduced by* Bernard J. Jandorf), *Toxicology Section, Medical Div., Army Chemical Center, Md.*
   Some properties of a transsulfurase responsbile for conversion of cyanide to thiocyanate.
2. **Otto Rosenthal,** *Harrison Dept. of Surgical Research, School of Medicine, University of Pennsylvania, Philadelphia.*
   The distribution of rhodanese.
3. **C. Boyd Shaffer and Frances H. Critchfield** (*introduced by* Leonard H. Cretcher), *Chemical Hygiene Fellowship, Mellon Inst., Pittsburgh*
   Lipotropic activity and toxicity of methoxinine (oxy-methionine).
4. **Herbert C. Tidwell** (*introduced by* Max N. Huffman), *Dept. of Biochemistry, Southwestern Medical College, Dallas*
   Some factors which influence methionine excretion in the rat.
5. **Cosmo G. Mackenzie, Julian R. Rachele** (*by invitation*), **Nancy Cross** (*by invitation*), **Joseph P. Chandler, and Vincent duVigneaud,** *Dept. of Biochemistry, Cornell University Medical College, New York*
   Study of the oxidation of the labile methyl group of dietary methionine traced with C^{14}.
6. **Jacob W. Dubnoff and Henry Borsook,** *William G. Kerckhoff Labs. of the Biological Sciences, California Inst. of Technology, Pasadena*
   Dimethylthetin as a methylator of homocysteine.
7. **Jakob A. Stekol,** *The Lankenau Hospital Research Inst., and the Inst. for Cancer Research, Philadelphia*
   Synthesis of S-benzyl-thiopyruvic acid and its conversion to N-acetyl-S-benzyl-L-cysteine in the rat.
8. **Kay Nakamura** (*by invitation*) **and Francis Binkley,** *Lab. for the Study of Hereditary and Metabolic Disorders and the Depts. of Biochemistry and Medicine, University of Utah School of Medicine, Salt Lake City*
   The enzymatic hydrolysis of glutathione.
9. **Julius Schultz** (*introduced by* Harry M. Vars), *Harrison Dept. of Surgical Research, School of Medicine, University of Pennsylvania, Philadelphia*
   Influence of the presence of a sterile abscess on the detoxication of brombenzene as mercapturic acid.

BIOCHEMISTRY A

Thursday, March 18, 1:45 p. m.

Room C, Convention Hall

Symposium on Hemin Pigments and Chromoproteins

David L. Drabkin, *Chairman*

1. **David L. Drabkin,** *University of Pennsylvania, Philadelphia*
   Distribution and metabolic aspects of derivatives of iron protoporphyrin (hemin).
2. **Paul F. Hahn,** *Meharry Medical College, Nashville*
   Metabolism of iron.
3. **W. Mansfield Clark,** *Johns Hopkins University, Baltimore*
   A systematic treatment of co-ordination complexes of iron protoporphyrins with nitrogenous bases.
4. **Jeffries Wyman, Jr.,** *Harvard University, Cambridge*
   The relation of physiological function and molecular structure in hemoglobin.
5. **Leonor Michaelis,** *The Rockefeller Inst. for Medical Research, New York*
   Molecular oxygen as a ligand in metal porphyrins and other metal complex compounds.

BIOCHEMISTRY BUSINESS MEETING

Thursday, March 18, 4:15 p.m.

Room C, Convention Hall

BIOCHEMISTRY B

Thursday, March 18, 1:45 p.m.

Room D, Convention Hall

Glycolysis

1. **Chalmers L. Gemmill,** *Dept. of Pharmacology, University of Virginia Medical School, Charlottesville*
   Inhibitory effects of naphthoquinones and related compounds on glycolysis. (Pharmacol.)
2. **Helena Gilder** (*introduced by* Vincent duVigneaud), **Marion H. Wilson** (*by invitation*), **and Johanna M. Lee** (*by invitation*), *Dept. of Biochemistry, Cornell University Medical College, New York*
   Studies on glycolysis of mouse lymphosarcoma.
3. **William H. Summerson, Helena Gilder** (*by invitation*), **and Johanna M. Lee** (*by invitation*), *Dept. of Biochemistry, Cornell University Medical College, New York*
   Effect of pH on the aerobic metabolism of lymphosarcoma cells.

4. **Robert M. Bird and John D. Evans** (*introduced by* William H. Summerson), *Dept. of Physiology, Cornell University Medical College, New York*
   Effect of pH on the aerobic metabolism of rabbit bone marrow.
5. **Marion K. Birmingham** (*by invitation*) **and K. A. C. Elliott,** *Montreal Neurological Inst., McGill University, Canada*
   Effects of pH and bicarbonate on brain tissue respiration and anaerobic glycolysis
6. **K. A. C. Elliott,** *Montreal Neurological Inst., McGill University, Canada*
   Metabolism of cerebral cortex from different areas and various animals and from epileptic patients.
7. **James A. Bain** (*by invitation*) **and J. Raymond Klein,** *Depts. of Pharmacology, Psychiatry, and Biological Chemistry, University of Illinois College of Medicine, Illinois Neuropsychiatric Inst., Chicago*
   Effect of carbon dioxide concentration on brain lactate.
8. **J. Raymond Klein and James A. Bain** (*by invitation*), *Depts. of Psychiatry, Biological Chemistry, and Pharmacology, University of Illinois College of Medicine, Illinois Neuropsychiatric Inst., Chicago*
   Effect of carbon dioxide concentration on the changes in brain metabolites accompanying convulsions.
9. **Winifred Ashby** (*introduced by* Joseph H. Roe), *Blackburn Lab., St. Elizabeths Hospital, Washington*
   Distribution pattern of carbonic anhydrase in the fetal central nervous system.

BIOCHEMISTRY BUSINESS MEETING

Thursday, March 18, 4:15 p.m.

ROOM C, CONVENTION HALL

BIOCHEMISTRY C

Thursday, March 18, 1:45 p.m.

ROOM B, CONVENTION HALL

Nutrition

1. **Robert A. Harte** (*by invitation*), **John J. Travers** (*by invitation*), **Peter Sarich** (*by invitation*), **and James B. Allison,** *Research Labs., Arlington Chemical Co., Yonkers*
   The effect of protein quality of previous intake on the consequences of acute starvation.
2. **James B. Allison, John A. Anderson** (*by invitation*), **and John I. White** (*by invitation*), *Bureau of Biological Research, Rutgers University, New Brunswick*
   Repletion of protein depleted dogs with whole egg and wheat gluten proteins.
3. **Bacon F. Chow and Shirley DeBiase** (*by invitation*), *Div. of Protein Chemistry, The Squibb Inst. for Medical Research, New Brunswick*
   Repletion of protein depleted dogs with various proteins and protein hydrolysates.
4. **Charles F. Kade, Jr. and Jessey Shepherd** (*introduced by* Aaron Arnold), *Biology Div., Sterling-Winthrop Research Inst., Rensselaer, N. Y.*
   The inhibitory effect of excess methionine on protein utilization. (Nutrition)
5. **Barnett Sure and Freeland Romans** (*by invitation*), *University of Arkansas, Fayetteville*
   Influence of the concentration of the vitamin B complex on protein utilization.
6. **Oliver H. Gaebler and Paul Bartlett** (*by invitation*), *Henry Ford Hospital, Detroit*
   Conversion of protein to glucose in vitamin deficiencies.
7. **Anthony A. Albanese, L. E. Holt, Jr., and Virginia I. Davis** (*by invitation*), **Selma E. Snyderman** (*by invitation*), **Marilyn Lein** (*by invitation*), **and Emilie M. Smetak** (*by invitation*), *New York University College of Medicine*
   The sulfur amino acid requirement of the infant.
8. **Wendell H. Griffith and Mary F. Nawrocki** (*by invitation*), *Dept. of Biological Chemistry, St. Louis University School of Medicine*
   The effect of threonine in choline deficiency. (Nutrition)
9. **W. W. Westerfeld and J. M. McKibbin** (*by invitation*), *Dept. of Biochemistry, Syracuse University College of Medicine, Syracuse, New York*
   Acetaldehyde metabolism in dogs maintained on a purified diet.

BIOCHEMISTRY BUSINESS MEETING*

Thursday, March 18, 4:15 p.m.

ROOM C, CONVENTION HALL

BIOCHEMISTRY A

Friday, March 19, 9:00 a.m.

ROOM C, CONVENTION HALL

Tricarboxylic Acid Cycle

1. **Murray Saffran and J. L. Prado** (*introduced by* K. A. C. Elliott), *Dept. of Biochemistry, McGill University, Montreal, Canada*
   Oxidation of members of the Krebs cycle by liver and kidney. Inhibition by *trans*-aconitate.

2. **B. L. Horecker and J. N. Stannard** (*by invitation*), *Lab. of Physical Biology, National Inst. of Health, Bethesda, Md.*
   The kinetics of inhibitor action in a carrier-linked system.
3. **D. T. Watts** (*introduced by* C. L. Gemmill), *Dept. of Pharmacology, University of Virginia Medical School, Charlottesville*
   Effect of some central nervous system stimulants and depressants on the activity of succinic dehydrogenase. (Pharmacol.)
4. **J. Leyden Webb** (*by invitation*), **Paul R. Saunders** (*by invitation*), **and Clinton H. Thienes,** *Dept. of Pharmacology and Toxicology, School of Medicine, University of Southern California, Los Angeles*
   Metabolism of rat heart slices. (Pharmacol.)
5. **O. N. Miller** (*by invitation*), **R. E. Olson** (*by invitation*), **and F. J. Stare,** *Dept. of Nutrition, Harvard University School of Public Health, and Dept. of Biological Chemistry, Harvard University Medical School, Boston*
   Utilization of pyruvate by heart ventricle *in vitro* as a function of time.
6. **R. E. Olson** (*by invitation*), **O. N. Miller** (*by invitation*), **and F. J. Stare,** *Dept. of Nutrition, Harvard University School of Public Health, and Dept. of Biological, Chemistry, Harvard University Medical School, Boston*
   Effect of biotin deficiency upon the respiration of cardiac muscle in ducklings.
7. **J. Ciethaml** (*by invitation*), **M. C. Gollub** (*by invitation*), **J. Speck** (*by invitation*), **and B. Vennesland,** *Dept. of Biochemistry, University of Chicago*
   Some properties of β-keto acid carboxylases from plants.
8. **F. Edmund Hunter, Jr.,** *Dept. of Pharmacology, Washington University School of Medicine, St. Louis*
   Phosphorylation of glucose due to a coupled oxidation-reduction between α-ketoglutaric acid and oxalacetic acid.
9. **Morris E. Friedkin** (*by invitation*) **and Albert L. Lehninger,** *Depts. of Biochemistry and Surgery, University of Chicago*
   Oxidation-coupled phosphate exchanges in the acid-insoluble esters of cell-free liver preparations.
10. **George Kalnitsky** (*introduced by* H. A. Mattill), *Dept. of Biochemistry, State University of Iowa, Iowa City*
    Citrate formation from oxalacetate.
11. **Samuel Natelson, Joseph B. Pincus, and Julius K. Lugovoy** (*introduced by* Albert E. Sobel), *Dept. of Biochemistry of the Jewish Hospital of Brooklyn*
    Response of citric acid levels to administration of glucose.

BIOCHEMISTRY B

Friday, March 19, 9:00 a.m.

Room D, Convention Hall

Proteolytic Enzymes

1. **Emil L. Smith,** *Lab. for the Study of Hereditary and Metabolic Disorders, and the Depts. of Biochemistry and Medicine, University of Utah School of Medicine, Salt Lake City*
   The hydrolysis of glycylglycine and glycyl-L-leucine by peptidases.
2. **H. Theo Hanson** (*by invitation*) **and Emil L. Smith,** *Lab. for the Study of Hereditary and Metabolic Disorders, and the Depts. of Biochemistry and Medicine, University of Utah School of Medicine, Salt Lake City*
   Action of peptidases on β-alanine peptides.
3. **C. E. Carter** (*introduced by* J. P. Greenstein), *Clinton National Lab., Oak Ridge, Tenn.*
   The metabolism of peptides of asparagine.
4. **Hans Neurath and Elaine Elkins** (*by invitation*), *Dept. of Biochemistry, Duke University School of Medicine, Durham*
   Kinetics and inhibition of carboxypeptidase activity.
5. **Helmut R. Gutmann** (*by invitation*) **and Joseph S. Fruton,** *Dept. of Physiological Chemistry, Yale Univ., New Haven*
   Partial purification of cathepsin II from swine kidney.
6. **Elizabeth K. Patterson, Marjorie E. Dackerman, and Jack Schultz** (*introduced by* Theodore F. Lavine), *Lankenau Hospital Research Inst. and Inst. for Cancer Research, Philadelphia*
   Peptidase increase during growth of the salivary glands of the fly, *Drosophila melanogaster.*
7. **Otto Schales, Regina M. Roux** (*by invitation*), **and Anne M. Suthon** (*by invitation*), *Chemical Research Lab. of the Alton Ochsner Medical Foundation, and Dept. of Biochemistry, Tulane University, New Orleans*
   Inhibition of peptic activity by hydrazine.
8. **Heinz Fraenkel-Conrat, R. S. Bean** (*by invitation*), **and Hans Lineweaver,** *Western Regional Research Lab., Albany, Calif., Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Dept. of Agriculture*
   Concerning the mechanism of interaction of egg white trypsin inhibitor and trypsin.
9. **Donald E. Bowman,** *Dept. of Biochemistry and Pharmacology, Indiana University School of Medicine, Indianapolis*
   Inactivation of trypsin by acetylation and iodination.

BIOCHEMISTRY C

Friday, March 19, 9:00 a.m.

ROOM B, CONVENTION HALL

Antibiotics and Bacterial Metabolism

1. **Ibert C. Wells** (*by invitation*), **William H. Elliott** (*by invitation*), **Sidney A. Thayer, and Edward A. Doisy,** *Lab. of Biological Chemistry, St. Louis University School of Medicine*
   Ozonolysis of the Pyo compounds.
2. **M. Adler** (*by invitation*) **and O. Wintersteiner,** *Div. of Organic Chemistry, The Squibb Inst. for Medical Research, New Brunswick*
   The penicillins produced by *Aspergillus flavus.*
3. **James D. Dutcher, O. Wintersteiner, and A. E. O. Menzel** (*by invitation*), *Div. of Organic Chemistry, The Squibb Inst. for Medical Research, New Brunswick*
   Structural investigation of hydroxyaspergillic acid, an antibiotic substance produced by *Aspergillus flavus.*
4. **Kent Wight** (*by invitation*) **and Dean Burk,** *National Cancer Inst., National Inst. of Health, Bethesda, Md.*
   The effect of streptomycin on deamination and oxygen consumption by resting cells of *E. coli.*
5. **Ralph W. McKee and Quentin M. Geiman** (*by invitation*), *Depts. of Biological Chemistry and of Comparative Pathology and Tropical Medicine, Harvard University Medical School, Boston*
   Methionine in the growth of the malarial parasite, *Plasmodium knowlesi.*
6. **G. M. Shull** (*by invitation*), **Richard W. Thomas** (*by invitation*), **and W. H. Peterson,** *Dept. of Biochemistry, University of Wisconsin, Madison*
   Nature of the "sporogenes vitamin" and nutrition of *Clostridium sporongenes*
7. **Gerrit Toennies and Dorothy L. Gallant** (*by invitation*), *Lankenau Hospital Research Inst. and Inst. for Cancer Research, Philadelphia*
   On the bacterial metabolism of lysine.
8. **Carl M. Lyman and K. A. Kuiken** (*by invitation*), *Dept. of Biochemistry and Nutrition, A and M College of Texas, College Station*
   Effect of vitamin B_6 on the utilization of *D* amino acids by lactic acid bacteria.
9. **Muriel M. Burr and W. A. Crandall** (*introduced by* L. I. Pugsley), *Food and Drugs Div., Dept. National Health and Welfare, Ottawa, Canada*
   A study of the microbiological assay of valine with estimation of precision of the method.

BIOCHEMISTRY D

Friday, March 19, 9:00 a.m.

ROOM S, CONVENTION HALL

Thyroid and Miscellaneous Papers

1. **Marschelle H. Power, William C. McConahey, Jr.** (*by invitation*), **F. Raymond Keating, Jr.** (*by invitation*), **and Joseph Berkson** (*by invitation*), *Divs. of Biochemistry, Medicine, and Biometry and Medical Statistics, Mayo Clinic, Rochester*
   Preliminary observations on the renal excretion of radioiodine after administration of tracer doses.
2. **DeWitt Stetten, Jr., and Adele Karp** (*by invitation*), *Dept. of Biochemistry, College of Physicians and Surgeons, Columbia University, New York*
   Influence of the thyroid incorporation of deuterium into tissue constituents of the rat.
3. **David L. Drabkin,** *Dept. of Physiological Chemistry, Graduate School of Medicine, University of Pennsylvania, Philadelphia*
   The effect of thyroidectomy and of thiouracil on cytochrome *C* metabolism and liver regeneration.
4. **Richard J. Winzler and Earl Frieden** (*by invitation*), *Dept. of Biochemistry and Nutrition, University of Southern California School of Medicine, Los Angeles*
   Thyroxine activity and antagonism of some structural analogues of thyroxine.
5. **J. F. McClendon, William C. Foster** (*by invitation*), **and J. W. Cavett** (*by invitation*), *Hahnemann Medical College, Philadelphia, and Salsbury's Labs., Charles City, Iowa*
   Pituitary-protein-bound iodine and the Plummer treatment of hyperthyroidism.
6. **Leonard T. Skeggs** (*by invitation*), **Jack R. Leonards** (*by invitation*), **and Victor C. Myers,** *Dept. of Clinical Biochemistry, Western Reserve University, Cleveland*
   The effect of hypertrophy on the chemical composition of rat cardiac muscle.
7. **Roger W. Marsters** (*by invitation*), **Jack R. Leonards** (*by invitation*), **and Victor C. Myers,** *Dept. of Clinical Biochemistry, Western Reserve University, Cleveland*
   The chemical composition of human aortas.
8. **Eugene Roberts** (*introduced by* Michael Somogyi), *Barnard Free Skin and Cancer Hospital, St. Louis*
   Determination of arginase activity in tissue homogenates: application to epidermal carcinogenesis in mice.
9. **F. B. Seibert, M. L. Pfaff** (*by invitation*), **and**

M. V. Seibert (*by invitation*), *Henry Phipps Inst., University of Pennsylvania, Philadelphia*

A quantitative method for a serum polysaccharide present in cases of tuberculosis and carcinoma.

BIOCHEMISTRY

Papers Read by Title

1. **Major Roberto Acosta,** *Mexican Army* (*by invitation*) **and Robert E. Johnson,** *U. S. Army Medical Nutrition Lab., Chicago*
   A micromodification of the Folin method for estimating urinary creatinine.
2. **James C. Andrews,** *Dept. of Biological Chemistry and Nutrition, School of Medicine, University of North Carolina, Chapel Hill*
   Partial hydrolysis products of human hair.
3. **Reginald M. Archibald and E. Stroh** (*by invitation*), *Dept. of Biochemistry, School of Hygiene and Public Health, Johns Hopkins University, Baltimore*
   Fractionation of urinary steroids by use of the Craig counter-current distribution machine.
4. **Howard H. Beard,** *Dept. of Biochemistry, Chicago Medical School*
   Correlation of the unitarian or trophoblastic thesis with the biological test of malignancy.
5. **Maurice Bruger, Samuel Member** (*by invitation*), **and Estelle Goldman** (*by invitation*), *Medical Research Lab., Dept. of Medicine, New York Post-Graduate Medical School and Hospital*
   The glucose content of the rat carcass.
6. **R. L. Dryer** (*by invitation*), **J. Katz** (*by invitation*), **W. D. Paul** (*by invitation*), **and J. I. Routh,** *Depts. of Biochemistry and Internal Medicine, College of Medicine, State University of Iowa, Iowa City*
   The anti-ulcer activity of aluminum dihyroxyaminoacetate.
7. **John D. Ferry and Sidney Shulman** (*by invitation*), *Dept. of Chemistry, University of Wisconsin, Madison*
   Influence of polyhydric alcohols on the clotting of fibrinogen.
8. **D. Breese Jones, Alvin Caldwell** (*by invitation*), **and Millard J. Horn** (*by invitation*), *Bureau of Human Nutrition and Home Economics, Agricultural Research Administration, U. S. Dept. of Agriculture, Washington*
   Availability of DL-lanthionine for the promotion of growth when added to a cystine-deficient diet.
9. **Harlan L. Klug** (*by invitation*), **George P. Lampson** (*by invitation*), **and Alvin L. Moxon,** *Chemistry Dept., South Dakota Agricultural Experiment Station, South Dakota State College, Brookings*
   Selenium tetraglutathione.
10. **Victor E. Levine and Sidney Merlis** (*by invitation*), *Dept. of Biological Chemistry and Nutrition, Creighton University School of Medicine, Omaha*
    Dibenzothiophene as a reagent for aldehydes, ketones and carbohydrates.
11. **Maurice V. L'Heureux** (*by invitation*), **Wilbur R. Tweedy, and Elinor M. Zorn** (*by invitation*), *Dept. of Biological Chemistry, Loyola University School of Medicine, Chicago*
    The excretion of labeled calcium by normal and thyroparathyroidectomized rats.
12. **George H. Mangun,** *Dept. of Labs., Henry Ford Hospital, Detroit*
    Effect of cytochrome C on the resistance of mice to anoxia.
13. **Mary E. Maver and Antoinette Greco** (*by invitation*), *National Cancer Inst., Bethesda, Md.*
    The hydrolysis of nuclear proteins by cathepsins. I. Calf thymus cathepsin.
14. **Dale K. Mecham** (*by invitation*) **and Harold S. Olcott,** *Western Regional Research Lab., Albany, Calif., Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Dept. of Agriculture*
    An egg yolk protein containing 10% phosphorus.
15. **Samuel Natelson** (*by invitation*), **Benjamin Kramer, and Marvin Sherman** (*by invitation*), *Pediatric Research Lab., Jewish Hospital of Brooklyn*
    Blood sugar changes following the administration of lactose in raw and evaporated milk.
16. **Joseph M. Quashnock** (*introduced by* William S. McEllroy), *Dept. of Physiological Chemistry, School of Medicine, University of Pittsburgh*
    Gas pressure regulation in flame photometry.
17. **G. G. Rudolph** (*by invitation*) **and L. T. Samuels,** *Dept. of Biochemistry, University of Utah School of Medicine, Salt Lake City*
    Early changes in the seminal vesicles of the castrate rat following administration of testosterone.
18. **Max Schlamowitz** (*by invitation*) **and David M. Greenberg,** *Div. of Biochemistry, University of California Medical School, Berkeley*
    Purification of phosphoglucomutase with "carbitol" acetate.
19. **Mona Spiegel-Adolf and Arnold S. J. Lee** (*by invitation*), *Dept. of Colloid Chemistry, Temple University School of Medicine, Philadelphia*
    Polarographic studies in cerebrospinal fluids.
20. **Mario Stefanini** (*introduced by* A. J. Quick),

Dept. of Internal Medicine, University of Roma, Italy, and Dept. of Biochemistry, Marquette University School of Medicine, Milwaukee

Mechanism of hyperbilirubinemia due to sodium nicotinate.

21. **E. L. R. Stokstad, J. Pierce** (*by invitation*), **T. H. Jukes, and A. L. Franklin** (*by invitation*), *Lederle Labs. Division, American Cyanamid Co., Pearl River, N. Y.*

The inhibition of pteroylglutamic acid conjugase by glutamic acid peptides of *p*-aminobenzoic acid.

22. **M. K. Walden** (*introduced by* A. K. Balls), *Enzyme Research Lab., Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Dept. of Agriculture, Albany, Calif.*

Serine content of purothionin.

AMERICAN SOCIETY FOR PHARMACOLOGY AND EXPERIMENTAL THERAPEUTICS, INC.

THIRTY-EIGHTH ANNUAL MEETING

PHARMACOLOGY A

Tuesday, March 16, 9:00 a.m.

ROOM E, CONVENTION HALL

Sympathetic and Related Agents

1. **Carlton C. Hunt** (*introduced by* McKeen Cattell), *Department of Pharmacology, Cornell University Medical College and Sloan-Kettering Institute, New York*

Structural relationship to sympatholytic activity of certain β-chlorethyl amines.

2. **Raymond P. Ahlquist,** *Department of Pharmacology, University of Georgia School of Medicine, Augusta*

Comparative effects of sympathomimetic amines on the vasomotor resistance of the kidney, mesentery and leg.

3. **Fred Shaffer** (*by invitation*), **P. L. Ewing and G. A. Emerson,** *Department of Pharmacology, University of Texas Medical Branch, Galveston*

Comparative effects of d-desoxyephedrine and the isomers of amphetamine on smooth muscle.

4. **Solomon Garb** (*by invitation*) **and Maynard B. Chenoweth,** *Dept. of Pharmacology, Cornell Univ. Medical College, New York*

Studies on ventricular fibrillation produced by epinephrine during hydrocarbon inhalation.

5. **James A. Richardson** (*by invitation*) **and R. P. Walton,** *Dept. of Pharmacology, Medical College of South Carolina*

Further analysis of the influence of autonomic innervation on drug responses of the heart and gut in unanesthetized dogs.

6. **Theodore O. King,** *Department of Pharmacology, Georgetown University School of Medicine, Washington*

The effect of pitressin on the hemodynamic responses of epinephrine and N-isopropyl-nor-epinephrine.

7. **William A. Wolff and Marina Hawkins** (*by invitation*), *Tobacco Research Laboratory, Bowman Gray School of Medicine, Wake Forest College, Winston-Salem*

A spectrophotometric method for nicotine in blood. (Biochem.)

8. **Amedeo S. Marrazzi,** *Department of Pharmacology and Therapeutics, Wayne University College of Medicine, Detroit*

Antagonism of nicotine and atropine.

9. **John B. Stanbury** (*introduced by* Otto Krayer), *Department of Pharmacology, Harvard Medical School*

The blocking action of magnesium ion on sympathetic ganglia.

10. **Clem A. Stone** (*by invitation*), **Paul Achenbach** (*by invitation*) **and Earl R. Loew,** *Dept. of Pharmacology, University of Illinois College of Medicine, Chicago*

Adrenergic blocking properties of certain halogenated ethylamine derivatives.

11. **J. A. Wells and David P. Rall** (*by invitation*), *Department of Pharmacology, Northwestern University Medical School, Chicago*

The influence of N-(2-bromoethyl)-N-ethyl-1-naphthalene-methylamine on the vasopressor response of a series of amines.

12. **B. Richards** (*by invitation*), **A. Cameron** (*by invitation*), **B. Craver, E. Herrold** (*by invitation*), *Division of Pharmacology, Research Department, Ciba Pharmaceutical Products, Inc., Summit, N. J.*

Detoxification of 2-benzyl-imidazoline hydrochloride (Priscol) by incubation with hepatic slices from the rat.

PHARMACOLOGY B

Tuesday, March 16, 9:00 a.m.

ROOM 9, CONVENTION HALL

General and Local Anesthetics

1. **Duncan E. Hutcheon** (*introduced by* G. H. W. Lucas), *Department of Pharmacology, University of Toronto*
   Sudden deaths during chloroform and cyclopropane anaesthesia.
2. **Barbara Rennick** (*by invitation*), **S. Donald Malton** (*by invitation*), **and Gordon K. Moe,** *Department of Pharmacology, the University of Michigan, Ann Arbor*
   The effect of cyclopropane on the work capacity of the dog heart.
3. **Charles H. Burnett** (*by invitation*), **Esther B. Gordon** (*by invitation*), **Gerald Shortz** (*by invitation*), **David W. Compton** (*by invitation*) **and Henry K. Beecher,** *Anesthesia Laboratory of the Harvard Medical School at the Massachusetts General Hospital and the Department of Medicine, Massachusetts General Hospital*
   A comparison of the effects of ether and cyclopropane on renal function in man.
4. **John C. Krantz, Jr. and C. Jelleff Carr,** *Department of Pharmacology, University of Maryland, School of Medicine, Baltimore*
   Anesthesia with cyclobutane.
5. **C. Jelleff Carr and John C. Krantz, Jr.,** *Department of Pharmacology, University of Maryland, School of Medicine, Baltimore*
   A comparative study of cyclic and noncyclic hydrocarbons on cardiac automaticity.
6. **H. R. Hulpieu and V. V. Cole,** *Department of Biochemistry and Pharmacology, Indiana University School of Medicine, Indianapolis*
   Procaine metabolism in dogs anesthetized by thiopental, ether and chloroform.
7. **Philip A. Lief** (*by invitation*), **Raymond Poet** (*by invitation*) **and Bernard B. Brodie,** *Departments of Anesthesiology and Biochemistry, New York University College of Medicine, and New York University Research Service, Goldwater Memorial Hospital, New York*
   The physiological disposition of procaine in man.
8. **Alexander G. Karczmar** (*by invitation*) **and Theodore Koppanyi,** *Department of Pharmacology and Materia Medica, Georgetown University School of Medicine, Washington*
   Action of central nervous system depressants at different growth periods of salamander (Amblystoma punctatum) larvae.
9. **Theodore Koppanyi and Alexander G. Karczmar** (*by invitation*), *Dept. of Pharmacology and Materia Medica, Georgetown University School of Medicine, Washington*
   Comparison of anesthetic action of acetanilid, tricaine (MS-222) and aliphatic depressants.

PHARMACOLOGY C

Tuesday, March 16, 9:00 a.m.

ROOM 1, CONVENTION HALL

Drug Toxicity

1. **R. A. Gardner** (*by invitation*), **P. L. Ewing, G. A. Emerson and A. E. Hansen,** *Departments of Pediatrics and Pharmacology, University of Texas Medical Branch, Galveston*
   Synergism in relation to toxicity of certain antiasthmatic drugs.
2. **J. K. Finnegan, P. S. Larson and H. B. Haag,** *Department of Pharmacology, Medical College of Virginia, Richmond*
   Comparative tolerance of dogs, cats and rabbits to nicotine.
3. **David I. Macht and Marcus Ostro** (*by invitation*), *Departments of Pharmacology and Radiology, Sinai Hospital, Baltimore*
   Phytotoxic effects of normal, pathological, and irradiated blood sera.
4. **Ruth Musser** (*introduced by* John C. Krantz, Jr.), *Department of Pharmacology, Univ. of Maryland, School of Medicine, Baltimore*
   Studies with the polyoxyethylene derivatives of sorbitan partial esters (Tweens).
5. **Alfred G. Lisi** (*introduced by* Charles M. Gruber), *Department of Pharmacology, Jefferson Medical College, Philadelphia*
   The toxicity of some surface active agents on the frog heart.
6. **Charles H. Hine and Herbert E. Christensen** (*by invitation*), *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco*
   The comparative activity of some α substituted glycerol and glycidyl ethers.
7. **Norman W. Karr and Edward L. Hendricks** (*introduced by* Norman A. David), *Department of Pharmacology, University of Oregon Medical School, Portland*
   Toxicity of intravenous ammonium salts.
8. **A. C. Conway** (*by invitation*), **F. S. Ting** (*by invitation*), **and Julius M. Coon,** *Department of Pharmacology, University of Chicago*

Effect of anti-cholinesterases upon procaine toxicity.

9. **O. Garth Fitzhugh, Arthur A. Nelson and Oma L. Holland** (*by invitation*), *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington*
The chronic toxicity of thiourea.
10. **Paul K. Smith, Abraham I. Gimble** (*by invitation*) **and Clarke Davison** (*by invitation*), *Department of Pharmacology, The George Washington University School of Medicine, Washington*
The tissue distribution and toxicity of emetine.
11. **Frieda M. Kunze** (*by invitation*) **and Edwin P. Laug,** *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington*
The penetration of lead through the skin of the rat.
12. **Frederick G. Germuth, Jr.** (*by invitation*) **and Harry Eagle,** *Laboratory of Experimental Therapeutics of the U. S. Public Health Service and the Johns Hopkins School of Hygiene and Public Health, Baltimore*
The efficacy of BAL (2,3-Dimercaptopropanol) in the treatment of experimental lead poisoning in rabbits.
13. **Lehman M. Lusky** (*by invitation*), **Herbert A. Braun and Edwin P. Laug,** *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington*
The protective action of BAL on experimental poisoning by lead, tungsten, copper and Paris green.

JOINT SESSION OF THE FEDERATION

Tuesday, March 16, 1:30 p.m.

Ballroom, Convention Hall

Program on page 314

PHARMACOLOGY BUSINESS MEETING

Tuesday, March 16, 4:15 p.m.

Room E, Convention Hall

PHARMACOLOGY

Tuesday, March 16, 7:30 p.m.

Ballroom, Convention Hall

Symposium on Human Pharmacological Experiments

1. History and Ethics of Human Physiological and Pharmacological Experiments.
Speaker, **A. C. Ivy.**
Discussant, **C. F. Schmidt.**
2. Autonomic Drugs Adaptable to Human Experimentation.
Speaker, **I. Starr.**
Discussant, **J. Comroe, Jr.**
3. Analgesic and Anesthetic Drugs in Man.
Speaker, **E. G. Gross.**
Discussant, **G. L. Maison.**
4. Miscellaneous Human Pharmacological Experiments.
Speaker, **K. Unna.**
Discussant, **H. Bruner.**
5. Dangers, Safeguards and Necessary Precautions.
General Discussion.

PHARMACOLOGY A

Wednesday, March 17, 9:00 a.m.

Room E, Convention Hall

Analgesics

1. **Robert A. Lehman, Herbert S. Kupperman** (*by invitation*) **and Joy Phillips** (*by invitation*), *Department of Therapeutics, New York University College of Medicine*
Studies on the relationship between chemical constitution and analgesic activity.
2. **Carl C. Pfeiffer, J. Santos-Martinez** (*by invitation*) **and Theodore R. Sherrod** (*by invitation*), *Department of Pharmacology, University of Illinois College of Medicine, Chicago*
The nature of the prosthetic groups of analgesics and their possible action as blocking agents.
3. **Nilkanth M. Phatak, James Maloney** (*by invitation*) **and Norman David,** *Department of Pharmacology, University of Oregon Medical School, Portland*
Use of hyperglycemic response for estimating addiction potentialities of analgesic compounds.
4. **F. P. Luduena, Lloyd C. Miller, Estelle Ananenko** (*by invitation*) **and J. D. Frick** (*by invitation*), *Biology Division, Sterling-Winthrop Research Institute, Rensselaer, N. Y.*
Some pharmacological actions of isomers of methadon.
5. **Elizabeth H. Jenney** (*by invitation*) **and Carl C. Pfeiffer,** *Department of Pharmacology, University of Illinois College of Medicine, Chicago*

Comparative analgesic and toxic effects of the optical isomers of methadon and isomethadon.

6. **Theodore R. Sherrod** (*by invitation*), **Robert Kaiser** (*by invitation*), **J. Santos-Martinez** (*by invitation*) **and Carl C. Pfeiffer,** *Department of Pharmacology, University of Illinois College of Medicine, Chicago*

   Methadon derivatives of pharmacological interest.

7. **James O. Hoppe** (*by invitation*) **and Lloyd C. Miller,** *Biology Division, Sterling-Winthrop Research Institute, Rensselaer, N. Y.*

   A comparison of the toxicity of methadon and some related compounds.

8. **Jane E. Denton** (*by invitation*), **Oliver H. Straus** (*by invitation*), **William E. Waddell** (*by invitation*) **and Henry K. Beecher,** *Anesthesia Laboratory of the Harvard Medical School at the Massachusetts General Hospital, Boston*

   A comparison of side actions and analgesic effects of morphine, amidone (4-4-diphenyl-6-dimethylamino-heptanone-3) and its isomers in man.

9. **Robert C. Batterman and Abraham M. Oshlag** (*by invitation*), *Department of Therapeutics, New York University College of Medicine*

   The effectiveness and toxicity of methadon.

10. **Marion Bell** (*by invitation*), **Joan E. Copeland** (*by invitation*) **and Eldon M. Boyd,** *Dept. of Pharmacology, Queen's Univ., Kingston, Canada*

    On the mechanism of the antitussive action of amidone.

11. **G. S. Eadie, F. Bernheim and D. B. Fitzgerald** (*by invitation*), *Dept. of Physiology and Pharmacology, Duke University School of Medicine, Durham*

    The effect of methadon on the cholinesterase of the brain.

12. **Ted A. Loomis** (*introduced by* James M. Dille), *Department of Pharmacology, University of Washington School of Medicine, Seattle*

    The response and the mechanism of the response of the duodenum to morphine.

13. **Sydney Ellis,** *Department of Physiology and Pharmacology, Duke University School of Medicine, Durham*

    Enzymic hydrolysis of morphine esters.

14. **Harris Isbell** (*by invitation*) **and A. J. Eiseman,** *Research Department, U. S. Public Health Service Hospital, Lexington, Ky.*

    Physical dependence liability of drugs of the methadon series and of 6-methyl-dihydromorphine. (Biochem.)

PHARMACOLOGY B

Wednesday, March 17, 9:00 a.m.

Room 9, Convention Hall

Antibiotics and Chemotherapeutic Agents

1. **R. J. Schachter** (*introduced by* N. Ercoli), *Department of Pharmacology and Chemotherapy, Warner Institute of Therapeutic Research, New York*

   The circulation of penicillin in the lymph.

2. **N. Ercoli, M. N. Lewis** (*by invitation*), **B. S. Schwartz** (*by invitation*), **M. H. Whitehead** (*by invitation*), *Dept. of Pharmacology and Chemotherapy, Warner Institute of Therapeutic Research, New York*

   The therapeutic significance of detectable penicillin levels.

3. **Harold M. Peck, Horace F. Russo, Elizabeth K. Tillson, William S. Waller** (*all by invitation*) **and Karl H. Beyer,** *Department of Pharmacology, The Medical Research Division, Sharp and Dohme, Inc., Glenolden, Pa.*

   The renal elimination of caronamide (4'-carboxyphenyl-methanesulfonanilide).

4. **Hans Molitor and Samuel Kuna** (*by invitation*), *Merck Institute for Therapeutic Research, Rahway*

   Pharmacologic studies of the neurotoxic properties of streptomycin.

5. **Joseph E. Hawkins, Jr. and Walter J. O'Shanny** (*by invitation*), *Merck Institute for Therapeutic Research, Rahway*

   Functional analysis of the chronic neurotoxic action of streptomycin.

6. **Lloyd D. Seager,** *Department of Pharmacology, The Woman's Medical College of Pennsylvania*

   The chemotherapeutic action of a number of furan derivatives.

7. **Andres Goth and Fabian J. Robinson** (*by invitation*), *Department of Physiology and Pharmacology, Southwestern Medical College, Dallas*

   Chemotherapeutic studies on a series of dithiocarbamates and their bismuth derivatives.

8. **David M. Tennent and Margaret L. Leland** (*introduced by* Randolph T. Major), *Merck Institute for Therapeutic Research, Rahway*

   A procedure for the determination of 4-aminosalicylic acid in blood and urine. (Biochem.)

9. **Les Loewe** (*by invitation*), **Albert Sobel, Oscar Gawron** (*by invitation*) **and Erna Alture-Werber** (*by invitation*), *Departments of Medicine and Biochemistry, Jewish Hospital of Brooklyn*

New penicillin products for prolonged blood levels. (Biochem.)

10. **John V. Scudi and Charles J. Duca** (*by invitation*), *Nepera Chemical Co., Inc., Yonkers*
Bacteriological characteristics of mandelamine. (Biochem.)

11. **R. M. Archibald and James R. Weisiger** (*by invitation, Hospital of The Rockefeller Institute for Medical Research, New York*
Reaction of plasmochin with formaldehyde. (Biochem.)

PHARMACOLOGY C

Wednesday, March 17, 9:00 a.m.

ROOM 1, CONVENTION HALL

Parasympathetic and Related Agents

1. **Clara Torda and Harold G. Wolff,** *New York Hospital and the Departments of Medicine (Neurology) and Psychiatry, Cornell University Medical College, New York*
Effect of convulsant and anticonvulsant agents on acetylcholine metabolism and effector organs acetylcholine sensitivity.

2. **K. R. Unna, K. Glaser** (*by invitation*), **E. Lipton** (*by invitation*), **and P. Patterson** (*by invitation*), *Departments of Pharmacology and Pediatrics, University of Illinois College of Medicine, Chicago*
Efficacy of atropine in children.

3. **Charles M. Gruber and Goldie Freedman Keyser** (*by invitation*), *Department of Pharmacology, Jefferson Medical College, Philadelphia*
The effects of 1-amino-1-phthalidylpropane hydrochloride on excised and intact intestine and uterus.

4. **Julius M. Coon and Paul R. Salerno** (*by invitation*), *University of Chicago Toxicity Laboratory and the Department of Pharmacology, University of Chicago*
A pharmacologic comparison of hexaethyl tetraphosphate (HETP) and tetraethyl pyrophosphate (TEPP) with physostigmine and neostigmine.

5. **H. Walter Jones, Jr.** (*by invitation*), **Bertram J. Meyer** (*by invitation*) **and Leonard Karel,** *Toxicology Section, Medical Division, Army Chemical Center, Md.*
The relationship of cholinesterase inhibiting activity to the acute toxicity of some organic phosphorus compounds.

6. **H. E. Himwich and A. M. Freedman** (*by invitation*), *Medical Division, Army Chemical Center, Md.*
Di-isopropyl fluorophosphate (DFP): Site of injection and variation in response. (Physiol.)

PHARMACOLOGY A

Wednesday, March 17, 1:45 p.m.

ROOM E, CONVENTION HALL

Curare and Anti-Curare-like Compounds

1. **K. K. Kimura** (*by invitation*) **and K. R. Unna,** *Department of Pharmacology, University of Illinois College of Medicine, Chicago*
Methods for the evaluation of compounds with curare-like action.

2. **Roger F. Varney** (*by invitation*), **Charles R. Linegar and Horace A. Holaday** (*by invitation*), *Biological and Chemical Laboratories, E. R. Squibb & Sons, New Brunswick*
The rabbit "Head-drop" method for the biological assay of curare and its alkaloids.

3. **E. F. Van Maanen** (*introduced by* Otto Krayer), *Department of Pharmacology, Harvard Medical School, Boston*
A comparison of curare alkaloids.

4. **E. L. McCawley,** *the Laboratories of Pharmacology and Toxicology and the Laboratory of Neuro Anatomy, Yale University School of Medicine, New Haven*
Some aspects of the action of curare on the central nervous system.

5. **G. M. Everett** (*introduced by* R. K. Richards), *Department of Pharmacology, Abbott Research Laboratories, North Chicago*
Studies on the toxicity and metabolism of d-tubocurarine.

6. **M. H. Pelletier** (*by invitation*) **and David Fielding Marsh,** *Department of Pharmacology, West Virginia University School of Medicine, Morgantown*
Synthetic curare compounds. II. d-N-Methyl-isochondrodendrine iodide and d-N-Methyl-O-methyl-isochondrodendrine iodide.

7. **David Fielding Marsh, Clark K. Sleeth** (*by invitation*), **and Eldon B. Tucker** (*by invitation*), *Departments of Pharmacology and Medicine, West Virginia University School of Medicine, Morgantown*
Synthetic curare compounds. III. d-N-Methyl-chondrodendrine iodide and d-N-Methyl-O-methyl-chondrodendrine iodide.

8. **John C. Burke** (*by invitation*) **and Charles R. Linegar,** *Pharmacological Development Division, E. R. Squibb & Sons, New Brunswick*
Pharmacological studies of myanesin and curare.

9. **H. A. Walker** (*by invitation*), **Arthur P. Richardson, P. Loeb** (*by invitation*) **and J. Perog** (*by invitation*), *Department of Pharmacology,*

Emory University School of Medicine, Emory University, Georgia, and Division of Pharmacology, Squibb Institute for Medical Research, New Brunswick

The paralytic and lethal action of myanesin, pentobarbital and combinations of these agents in mice.

10. **James L. Morrison, Arthur P. Richardson and Harry A. Walker** (*by invitation*), *Department of Pharmacology, Emory University, Georgia*

Rate of disappearance from the blood and distribution in tissues of 3-(Orthotoloxyl)-1,2-propanediol (Myanesin).

11. **Mitchell Zweig** (*by invitation*), **F. Steigmann, and Karl A. Meyer** (*by invitation*), *Hektoen Institute for Medical Research of Cook County Hospital and the Department of Surgery, Northwestern University School of Medicine*

The effect of tetraethyl ammonium chloride on gastric secretion and motility.

12. **Charles J. Kensler** (*introduced by* McKeen Cattell), *Department of Pharmacology, Cornell University Medical College*

The anticurare activity of congo red and related compounds.

PHARMACOLOGY B

Wednesday, March 17, 1:45 p.m.

Room D, Convention Hall

Joint Session, Biometrics Section, American Statistical Association, and the Pharmacological Society

1. **E. M. Jellinek** (*introduced by* C. I. Bliss), *Department of Applied Physiology, Yale University, New Haven*

Experimental bromism in man.

2. **Graham Chen, A. C. Bratton, Jr. and Charles Ensor** (*by invitation*), *Research Laboratories, Parke, Davis & Company, Detroit*

The joint antihistaminic effect and acute toxicity of adrenalin and benadryl HCl.

3. **Rex G. Fluharty** (*introduced by* Lloyd C. Miller), *Radioactivity Center, Massachusetts Institute of Technology*

Statistical problems involved in the use of radioactive tracers in pharmacology.

4. **J. T. Litchfield, Jr., and F. Wilcoxon** (*by invitation*), *Chemotherapy Division, Stamford Research Laboratories, American Cyanamid Company, Stamford*

A simplified method of evaluating dose-effect experiments.

5. **R. H. Noel and M. A. Brumbaugh** (*introduced by* Lloyd C. Miller), *Bristol Laboratories, Inc., Syracuse*

Some applications of sequential analysis.

Federation "Mixer"

Wednesday, March 17, 9:00 p.m.

Arena, Convention Hall

PHARMACOLOGY A

Thursday, March 18, 9:00 a.m.

Room E, Convention Hall

Barbiturates and Analgesics

1. **Walter M. Booker, David M. French, Pedro A. Molano, and Cecil Rhodes** (*introduced by* A. H. Maloney), *Department of Pharmacology, College of Medicine, Howard University, Washington*

Re-evaluation of the effectiveness of metrazol as an analeptic agent in barbiturate depression.

2. **V. V. Cole, S. H. Hopper** (*by invitation*), **and H. R. Hulpieu,** *Depts. of Pharmacology and Public Health, Indiana Univ. School of Medicine, Indianapolis*

Effects of surface active agents on the activity of pentobarbital and barbital.

3. **Marshall R. Warren** (*by invitation*), **Charles R. Thompson** (*by invitation*), **and Harold W. Werner,** *Pharmacology Department, Research Labs., The Wm. S. Merrell Co., Cincinnati*

Hypnotic properties and toxicity of 2-ethyl-3-propylglycidamide.

4. **A. R. Kelly** (*by invitation*), **F. E. Shideman and B. J. Adams** (*by invitation*), *Dept. of Pharmacology, University of Michigan, Ann Arbor*

Comparison of blood levels of thiopental (Pentothal), 5-allyl-5-(1-methylbutyl)-2-thiobarbituric acid (Surital) and thioethamyl in the dog.

5. **F. E. Shideman, A. R. Kelly** (*by invitation*), **L. E. Lee** (*by invitation*), **V. F. Lowell** (*by invitation*) **and B. J. Adams** (*by invitation*), *Department of Pharmacology, University of Michigan and Ypsilanti State Hospital, Ypsilanti, Mich.*

The effect of hepatic dysfunction in man on the duration of action of thiopental (Pentothal).

6. **Leo R. Goldbaum** (*by invitation*) **and Paul K. Smith,** *Department of Pharmacology, George Washington University School of Medicine, Washington*

The binding of barbiturates by human and bovine serum albumin.

7. **W. J. R. Camp, V. A. Gant** (*by invitation*) **and John F. Polli** (*by invitation*), *Dept. of Toxicology and Pharmacology, University of Illinois College of Medicine, Chicago*

The use of ultraviolet rays in a screening test

for some barbiturates and in reading Gutzeit papers.

8. **D. D. Bonnycastle and Jacob Molland** (*by invitation*), *Departments of Pharmacology, Yale School of Medicine, and University of Oslo, Norway*
An examination of potentiating action of aspirin upon codeine in raising the pain threshold.

PHARMACOLOGY B

Thursday, March 18, 9:00 a.m.

ROOM 9, CONVENTION HALL

Chemotherapeutic Agents

1. **Thomas H. Maren and Gilbert F. Otto** (*introduced by* E. K. Marshall, Jr.), *Department of Pharmacology, Johns Hopkins University School of Medicine and Department of Parasitology, Johns Hopkins School of Hygiene and Public Health, Baltimore*
The distribution, excretion, and blood levels of antimony following administration of antimonials to various mammalian species.
2. **Ralph G. Smith, James Y. P. Chen** (*by invitation*) **and Marguerite Magee** (*by invitation*), *Department of Pharmacology, Tulane University School of Medicine, New Orleans*
The distribution and excretion of antimony following administration of antimony potassium tartrate and neostibosan.
3. **Maxwell Schubert and Evelyn Goldberg** (*by invitation*), *Department of Therapeutics, New York University College of Medicine*
Comparison of therapeutic values of several antimonials in experimental schistosomiasis mansoni in mice.
4. **Harold N. Wright, Elizabeth M. Cranston, Wayne A. Chadbourn** (*by invitation*), **Ashton C. Cuckler** (*by invitation*), **Dominic DeGuisti** (*by invitation*) **and Raymond N. Bieter,** *Department of Pharmacology, University of Minnesota Medical School, Minneapolis*
Rosaniline base (CI 677) as a prophylactic in schistosoma mansoni infections in mice.
5. **Clara S. Genther** (*by invitation*), **Wanda Squires** (*by invitation*), **Rochelle Fradkin** (*by invitation*), **and L. H. Schmidt,** *Christ Hospital Institute of Medical Research, Cincinnati*
Malaria chemotherapy: 1. The response of trophozoite-induced infections with *Plasmodium cynomolgi* to various antimalarial drugs.
6. **L. H. Schmidt, Rochelle Fradkin** (*by invitation*), **Wanda Squires** (*by invitation*), **and Clara S. Genther** (*by invitation*), *Christ Hospital Institute of Medical Research, Cincinnati*
Malaria chemotherapy: 2. The response of sporozoite-induced infections with *Plasmodium cynomolgi* to various antimalarial Drugs.
7. **David Lehr,** *Department of Pharmacology, New York Medical College, Flower and Fifth Avenue Hospitals, New York*
Low toxicity and high therapeutic efficacy of combined sulfonamides.
8. **Celia White Tabor** (*by invitation*), **Judith Baily** (*by invitation*) **and Paul K. Smith,** *Department of Pharmacology, George Washington University School of Medicine, Washington*
Some metabolic products of para-aminobenzoic acid.
9. **E. Leong Way, Rowena Weiss** (*by invitation*), **Donald L. Howie** (*by invitation*) **and Paul K. Smith,** *Department of Pharmacology, George Washington University School of Medicine, Washington*
The fate of p-aminosalicylic acid in the animal body.
10. **W. T. McClosky, M. I. Smith and J. E. G. Frias** (*by invitation*), *Division of Physiology, National Institute of Health, Bethesda, Md.*
The action of p-aminosalicylic acid (PAS) in experimental tuberculosis.
11. **S. H. Hopper** (*by invitation*), **V. V. Cole, H. R. Hulpieu, and H. A. Raidt** (*by invitation*), *Departments of Public Health, Pharmacology, and Microbiology, Indiana University School of Medicine, Indianapolis*
Opposite effects in vitro and in vivo of a surface active agent on mycobacterium tuberculosis.

PHARMACOLOGY C

Thursday, March 18, 9:00 a.m.

ROOM 1, CONVENTION HALL

Agents Affecting the Blood and Vascular System

1. **Anthony M. Ambrose,** *Pharmacology Division, Bureau of Agricultural and Industrial Chemistry, U. S. Department of Agriculture, Albany*
The effect of rutin on blood pressure in dogs and rabbits.
2. **Walter Freyburger** (*by invitation*), **Luis R. Capo** (*by invitation*), **and Gordon K. Moe,** *Department of Pharmacology, the University of Michigan, Ann Arbor*
The effect of tetraethylammonium on the pressor response to anoxia and asphyxia.
3. **Donald C. Kunze** (*by invitation*), **J. Richard R. Bobb** (*by invitation*) **and Harold D. Green,** *Department of Physiology and Pharmacology,*

Bowman Gray School of Medicine of Wake Forest College, Winston-Salem

Vasomotor activity of NU-1683 and other drugs upon the rat meso-appendix.

4. **D. M. Green and M. Glover** (*by invitation*), *School of Medicine, University of Washington, Seattle*

   Factors influencing the hypertensive action of desoxycorticosterone.

5. **Joseph G. Bird** (*introduced by* John C. Krantz, Jr.), *Department of Pharmacology, University of Maryland School of Medicine, Baltimore*

   Prolonged depressor effect of water soluble organic nitrates.

6. **Albert J. Begany** (*by invitation*) **and Joseph Seifter,** *Wyeth Institute of Applied Biochemistry, Philadelphia*

   Studies on a heparin-like compound, algarin.

7. **David W. Fassett and E. Sterling Nichol** (*by invitation*), *Cardiology Service, James M. Jackson Memorial Hospital, Miami*

   Absorption and potency of small divided doses of dicumarol in man.

8. **Rose Marie Carlson** (*by invitation*) **and Lloyd D. Seager,** *Department of Pharmacology, Womans Medical College of Pennsylvania, Philadelphia*

   The acute toxic effects of dicumarol.

9. **Robert V. Brown,** *Division of Pharmacology, University of Tennessee, Memphis*

   Quantitative epinephrine pressor effects. (Physiol.)

10. **E. William Ligon, Jr.** (*by invitation*), **Robert A. Madden** (*by invitation*), **Paul L. Davis** (*by invitation*) **and Paul K. Smith,** *Department of Pharmacology, George Washington University School of Medcine, Washington*

    Absorption of salicylates from intestinal loops in the dog.

11. **Bernard B. Brodie and Julius Axelrod** (*by invitation*), *Department of Biochemistry, New York University College of Medicine, and New York University Research Service, Goldwater Memorial Hospital, and Laboratory of Industrial Hygiene, New York*

    The physiological disposition of acetophenetidin (p-ethoxyacetanilide) in man.

12. **R. R. Sonnenschein, R. Jamison, L. Lovseth, W. Cassels** (*by invitation*) **and A. C. Ivy,** *Departments of Clinical Science and Anesthesia, University of Illinois College of Medicine, Chicago*

    The mechanism of nitrous oxide analgesia. (Physiol.)

PHARMACOLOGY A

Thursday, March 18, 1:45 p.m.

ROOM E, CONVENTION HALL

Cardiac and Related Agents

1. **R. P. Walton, M. F. Patton** (*by invitation*), **H. P. Jones** (*by invitation*) **and J. S. Leary** (*by invitation*), *Department of Pharmacology, Medical College of South Carolina, Charleston*

   Comparative increase in ventricular contractile force produced by several cardiac glycosides.

2. **Mark Nickerson and George M. Nomaguchi** (*by invitation*), *Department of Pharmacology, University of Utah School of Medicine, Salt Lake City*

   Dibenamine protection against cyclopropane-epinephrine cardiac arrhythmias.

3. **K. I. Melville,** *Dept. of Pharmacology, McGill University, Montreal*

   The anti-fibrillation action of papaverine and its value in cardiac resuscitation after chloroform-adrenaline ventricular fibrillation.

4. **Otto Krayer and Alfred Farah,** *Department of Pharmacology, Harvard Medical School, Boston*

   Action of cysteine and of dimercaptopropanol in heart failure caused by sodium bismuth tartrate.

5. **N. J. Giarman** (*introduced by* E. L. McCawley), *Laboratories of Pharmacology and Toxicology, Yale University School of Medicine, New Haven*

   Toxicity and cardiotonic activity of antibiotic lactones and synthetic analogs on the isolated frog heart.

6. **A. Cameron** (*by invitation*), **M. Laskey** (*by invitation*), **B. Craver,** *Division of Pharmacology, Research Department, Ciba Pharmaceutical Products, Inc., Summit, N. J.*

   Actions and minimally effective doses of 17 common drugs on perfused hearts of 5 species.

7. **Maynard B. Chenoweth and Solomon Garb** (*by invitation*), *Department of Pharmacology, Cornell Univ. Medical College, New York, N. Y.*

   The electrogram of the papillary muscle.

8. **A. Farah and E. Szmuszkoviez** (*introduced by* James M. Dille), *Departments of Pharmacology, University of Washington Medical School, Seattle, and the American University of Beirut, Lebanon*

   Some factors influencing the activity of cardiac glycosides in the rat.

PHARMACOLOGY BUSINESS MEETING

Thursday, March 18, 4:15 p.m.

ROOM E, CONVENTION HALL

PHARMACOLOGY DINNER

Thursday, March 18, 7:00 p.m.

HOTEL HADDON HALL

PHARMACOLOGY B

Thursday, March 18, 1:45 p.m.

ROOM 9, CONVENTION HALL

1. **Salab A. Abdel Tawab** (*introduced by* John C. Krantz, Jr.), *Department of Pharmacology, University of Maryland School of Medicine, Baltimore*
   The pharmacology of organic thiocyanates.
2. **Ewart A. Swinyard** (*by invitation*) **and James E. P. Toman,** *Departments of Pharmacology, Pharmacy, and Physiology, University of Utah, Salt Lake City*
   Effects of alterations in body temperature on properties of convulsive seizures in rats.
3. **Avram Goldstein** (*introduced by* Otto Krayer), *Department of Pharmacology, Harvard Medical School, Boston*
   Inhibitors of purified human plasma cholinesterase.
4. **Rafael Mendez and Ernesto Sodi** (*by invitation*), *Department of Physiology and Pharmacology of the National Institute of Cardiology, Mexico City*
   The effect of some glycerin esters upon the blood pressure, uterus, and respiration.
5. **E. Herrold** (*by invitation*), **H. Hays** (*by invitation*), **G. Holmquist** (*by invitation*), **B. Richards** (*by invitation*), **E. Oppenheimer,** *Division of Pharmacology, Research Department, Ciba Pharmaceutical Products, Inc., Summit, N. J.*
   The water-soluble I-glucoside of desoxycorticosterone acetate; its adrenal cortical activity in adrenalectomized dogs.
6. **Victor A. Drill and B. Frame** (*by invitation*), *Department of Pharmacology, Yale University School of Medicine, New Haven*
   Antidiuretic activity of liver extracts and of urine from patients with hepatic cirrhosis.
7. **Douglas S. Riggs** (*introduced by* Otto Krayer), *Department of Pharmacology, Harvard Medical School and The Thyroid Clinic, Massachusetts General Hospital*
   Elevation of serum protein-bound iodine after large doses of radio-active iodine.
8. **J. K. W. Ferguson and E. A. Sellers,** *Department of Pharmacology, University of Toronto*
   Effect of various compounds containing iodine and other halogens on thiouracil goitre.
9. **D. G. Wenzel** (*by invitation*), **O. S. Orth, R. T. Capps** (*by invitation*), **and A. H. Uhl** (*by invitation*), *Departments of Pharmacy and Pharmacology, University of Wisconsin, Madison*
   Actions of podophyllum derivatives.
10. **Allan D. Bass,** *Pharmacology Department, Syracuse University*
    Response of experimental lymphoma to certain organic sulfides, sulfones and chlorides.

PHARMACOLOGY BUSINESS MEETING

Thursday, March 18, 4:15 p.m.

ROOM E, CONVENTION HALL

PHARMACOLOGY DINNER

Thursday, March 18, 7:00 p.m.

HOTEL HADDON HALL

PHARMACOLOGY C

Thursday, March 18, 1:45 p.m.

ROOM 1, CONVENTION HALL

Drug Toxicity

1. **Geoffrey Woodward** (*by invitation*), **Bernard Davidow** (*by invitation*), **and Arthur A. Nelson,** *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington*
   Effects observed in dogs following the prolonged feeding of DDT and its analogues.
2. **Lloyd W. Hazelton and Emily Godfrey** (*by invitation*), *Hazelton Laboratories, Falls Church, Va.*
   Pharmacological actions of o,o-diethyl o-p-nitrophenyl thiophosphate.
3. **Ernest C. Hagan and Geoffrey Woodard** (*introduced by* Arnold J. Lehman), *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington*
   Toxicity of o,o-diethyl o-p-nitrophenyl thiophosphate (Parathion).
4. **Kenneth P. DuBois, John Doull** (*by invitation*), **and Julius M. Coon,** *University of Chicago Toxicity Laboratory and the Department of Pharmacology, University of Chicago*
   Toxicity and mechanism of action of p-nitropheny -diethyl-thionophosphate (E605).

5. **Jack L. Radomski, Geoffrey Woodard, Carter D. Johnston** (*introduced by* Arnold J. Lehman), *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington*
   Toxicological properties of the causative agent of canine hysteria.
6. **W. F. von Oettingen, C. C. Powell** (*by invitation*), **N. E. Sharpless** (*by invitation*) **and L. J. Pecora** (*by invitation*), *Laboratory of Physical Biology, National Institute of Health, Bethesda, Md.*
   Comparative studies of the toxicity and physiological action of chlorinated methanes with reference to their physical and chemical characteristics.
7. **Edwin P. Laug,** *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington*
   Tissue distribution of a toxicant following oral ingestion of benzene hexachloride by rats.
8. **Bernard Davidow, Ernest C. Hagan and Geoffrey Woodard** (*introduced by* Arnold J. Lehman), *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington*
   A comparison of the fat storage of the isomers of benzene hexachloride in rats.
9. **Leonard Karel,** *Toxicology Section, Medical Division, Army Chemical Center, Md.*
   A potential rodenticide—"Fanyline."
10. **Elliott A. Maynard, William L. Downs, and Harold C. Hodge** (*introduced by* Harvey B. Haag), *Department of Pharmacology, University of Rochester, N. Y.*
    Preliminary data on rat feeding with beryllium.
11. **Harold C. Hodge, Elliott A. Maynard and William L. Downs** (*introduced by* Harvey B. Haag), *Department of Pharmacology, University of Rochester, N. Y.*
    Certain aspects of the acute toxicity of beryllium following intraperitoneal injection.
12. **Charles W. LaBelle and Martha Reid Cucci** (*introduced by* Harvey B. Haag), *Department of Pharmacology, University of Rochester, N. Y.*
    Preliminary studies in the toxicity of beryllium: The effect of intratracheal injections in experimental animals.
13. **George F. Sprague, Alton G. Pettengill and Herbert E. Stokinger** (*introduced by* Harvey B. Haag), *Department of Pharmacology, University of Rochester, N. Y.*
    Initial studies of the inhalation toxicity of beryllium sulfate.
14. **Sidney Laskin, Robert A. N. Turner and Herbert E. Stokinger** (*introduced by* Harvey B. Haag), *Department of Pharmacology, University of Rochester, N. Y.*
    Analysis of dust and fume hazards in a beryllium plant.

PHARMACOLOGY BUSINESS MEETING

Thursday, March 18, 4:15 p.m.

Room E, Convention Hall

AMERICAN SOCIETY FOR PHARMACOLOGY AND EXPERIMENTAL THERAPEUTICS

Annual Dinner

Thursday, March 18, 7:00 p.m.

Hotel Haddon Hall

PHARMACOLOGY

Motion Pictures

Thursday, March 18, 8:00 p.m.

Room C, Convention Hall

Program on page 314

PHARMACOLOGY A

Friday, March 19, 9:00 p.m.

Room E, Convention Hall

Histamine, Anti-histamine and Spasmolytic Agents

1. **Donald L. Cook** (*by invitation*), **W. E. Hambourger and Martin M. Winbury** (*by invitation*), *Pharmacology Laboratories, Research Department, G. D. Searle & Co., Chicago*
   Inhibition of histamine hypotension by certain heterocyclic substituted ethylamines in the cat.
2. **Homer B. Freese** (*by invitation*), **W. E. Hambourger and Patricia M. Michiels** (*by invitation*), *Pharmacology Laboratories, Research Department, G. D. Searle & Co., Chicago*
   The effectiveness of certain substituted ethylamines against histamine spray-induced fatality in the guinea pig.
3. **Julio C. Castillo** (*introduced by* Edwin J. de Beer), *The Wellcome Research Laboratories, Tuckahoe*
   The anaphylactic guinea pig trachea and its response to antihistamine and bronchodilator drugs.

4. **J. Smith** (*by invitation*), **A. Cameron** (*by invitation*), **B. Craver,** *Division of Pharmacology, Research Department, Ciba Pharmaceutical Products, Inc., Summit, N. J.*
   Method for kymographic recording of CSF pressure; effects of histamine, antistine, PBZ, et. al., thereon.
5. **Herbert Tabor and Sanford M. Rosenthal,** *Division of Physiology, National Institute of Health, Bethesda, Md.*
   Urinary excretion of histamine following oral administration, employing an improved colorimetric method.
6. **Lillian Alonso** (*by invitation*), **Maxine Adams** (*by invitation*), **Louise Goddard** (*by invitation*), **Marion Jaeger** (*by invitation*), **and J. T. Litchfield, Jr.,** *Chemotherapy Division, Stamford Research Laboratories, American Cyanamid Company, Stamford*
   Mode of action of antihistaminic agents.
7. **George W. Stavraky,** *Department of Physiology, Faculty of Medicine, University of Western Ontario, London, Canada*
   Potentiation of the effect of antihistaminic agents by iron compounds.
8. **Annette LaBelle and Richard Tislow** (*introduced by* O. Krayer), *Biological Research Laboratories, Schering Corporation, Bloomfield*
   Pharmacological properties of trimeton, a new antihistaminic compound.
9. **B. Craver, W. Barrett** (*by invitation*), **A. Cameron** (*by invitation*), **H. Hays** (*by invitation*), **G. Holmquist** (*by invitation*), **A. Mackenzie** (*by invitation*), **J. Smith** (*by invitation*), *Division of Pharmacology, Research Department, Ciba Pharmaceutical Products, Inc., Summit, N. J.*
   Pharmacological actions of 2-phenylbenzyl-aminomethyl-imidazoline (Antistine).
10. **Robert H. Dreisbach and Nai Chu** (Fellow, American Bureau for Medical Aid to China Inc.; *by invitation*), *Dept. of Pharmacology and Therapeutics, Stanford University School of Medicine, San Francisco*
    Procaine as an antiallergic agent.
11. **A. J. Glazko, W. A. Dill** (*by invitation*), **and D. A. McGinty,** *Research Laboratories, Parke, Davis and Co., Detroit*
    Distribution and excretion of benadryl (β-dimethylaminoethyl benzhydryl ether). (Biochem.)
12. **T. C. Barnes,** *Department of Pharmacology, Hahnemann Medical College and Hospital of Philadelphia*
    The adrenergic properties of antihistaminic drugs determined by electrical measurements. (Physiol.)

PHARMACOLOGY B

Friday, March 19, 9:00 a.m.

Room 9, Convention Hall

Cardiac and Related Agents

1. **Elwood L. Foltz, Sau Ki Wong and James E. Eckenhoff** (*introduced by* Carl F. Schmidt), *Department of Pharmacology, School of Medicine, University of Pennsylvania, Philadelphia*
   Effects of certain "cardiac stimulant" drugs on coronary circulation and cardiac oxygen metabolism.
2. **Murray Finkelstein** (*by invitation*) **and Oscar Bodansky,** *Dept. of Pharmacology, Cornell University Medical College, New York*
   Effect of scilliroside on the respiration of cat heart muscle.
3. **Albert Wollenberger** (*introduced by* Otto Krayer), *Harvard Medical School, Boston*
   Effect of ouabain and digoxin on the energy-rich phosphate store of the heart.
4. **Paul R. Saunders** (*by invitation*), **J. Leyden Webb** (*by invitation*), **and Clinton H. Thienes,** *Department of Pharmacology and Toxicology, School of Medicine, University of Southern California, Los Angeles*
   Effect of quinidine on the metabolism of the rat heart.
5. **Harry K. Iwamoto and Frederick K. Bell,** (*introduced by* John C. Krantz, Jr.), *Department of Pharmacology, School of Medicine, Univ. of Maryland, Baltimore*
   The Baljet reaction and pharmacodynamic studies of diginin and gitogenin.
6. **Walter Modell, Nathaniel T. Kwit** (*by invitation*), **Conrado Dayrit** (*by invitation*), **S. J. Shane** (*by invitation*) **and Harry Gold,** *Department of Pharmacology of Cornell University Medical College, Cardiovascular Research Unit of the Beth Israel Hospital, and Cardiac Service of the Hospital for Joint Diseases, New York*
   Comparison of digitalis glycosides with their genins in man.
7. **Nathaniel T. Kwit** (*by invitation*), **Walter Modell, Lawrence W. Hanlon** (*by invitation*), **Joseph G. Benton** (*by invitation*), **Elaine W. Cotlove** (*by invitation*), **Morris Pearlmutter** (*by invitation*), **Sidney M. Greenberg** (*by invitation*), **Milton L. Kramer** (*by invitation*), **and Harry Gold,** *Department of Pharmacology of Cornell University Medical College, Cardiovascular Research Unit of the Beth Israel Hospital, and Cardiac Service of the Hospital for Joint Diseases, New York*

The daily dose of the mercurial diuretic and the maintenance dose to control congestive failure.

8. **S. J. Shane** (*by invitation*), **Conrado Dayrit** (*by invitation*), **Joseph G. Benton** (*by invitation*), **Elaine W. Cotlove** (*by invitation*), **Lawrence W. Hanlon** (*by invitation*), **Walter Modell, and Harry Gold,** *Department of Pharmacology of Cornell University Medical College, Cardiovascular Research Unit of the Beth Israel Hospital, and Cardiac Service of the Hospital for Joint Diseases, New York*

Onset and duration of action of quinidine in the heart after oral administration in man.

9. **Conrado Dayrit** (*by invitation*), **Eduardo Faraco** (*by invitation*), **Nathaniel T. Kwit** (*by invitation*), **Walter Modell, and Harry Gold,** *Department of Pharmacology of Cornell University Medical College, Cardiovascular Research Unit of the Beth Israel Hospital, and Cardiac Service of the Hospital for Joint Diseases, New York*

Speed of action of thevetin after intravenous injection in man.

PHARMACOLOGY C

Friday, March 19, 9:00 a.m.

ROOM 1, CONVENTION HALL

1. **R. M. Whitrock** (*by invitation*), **H. L. Tieche** (*by invitation*), **and M. H. Seevers,** *Department of Pharmacology, University of Michigan, Ann Arbor*

The effect of "denervation" on the response of intestinal fistulae to several drugs.

2. **Ralph Uber** (*by invitation*), **Patricia Collins Cowdery** (*by invitation*) **and George E. Farrar, Jr.,** *Department of Medicine, Temple University School of Medicine, Philadelphia*

The spreading of Evan's blue injected intradermally.

3. **Ralph W. Brauer and Rita L. Pessotti** (*introduced by* Chapman Reynolds), *Department of Pharmacology and Experimental Therapeutics, School of Medicine, Louisiana State University, New Orleans*

The mechanism of the extraction of bromosulfalein from blood plasma by the liver.

4. **H. Ward Smith** (*introduced by* G. H. W. Lucas), *Department of Pharmacology, University of Toronto*

The specificity of the determination of alcohol in biological fluids.

5. **Edward B. Truitt, Jr.** (*introduced by* John C. Krantz, Jr.), *Department of Pharmacology, University of Maryland, School of Medicine, Baltimore*

The quantitative estimation of theophylline in blood.

6. **R. A. Woodbury, George P. Child** (*by invitation*), **and R. Torpin** (*by invitation*), *Division of Pharmacology, University of Tennessee, Memphis, and Departments of Pharmacology and Obstetrics and Gynecology, University of Georgia School of Medicine, Augusta*

Oxytocic drugs and dysmenorrhea.

7. **Joseph Thomas Roberts,** *University of Arkansas School of Medicine, Little Rock, Arkansas and Gallinger Municipal Hospital, Washington*

The effect of globulin insulin and protamine zinc insulin on the diurnal blood sugar curve and the daily glycosuria of diabetics, and the clinical use of globin insulin.

8. **A. J. Plummer** (*introduced by* George L. Maison), *Department of Pharmacology, Boston University School of Medicine*

Blood and urine levels of theophylline after intravenous injection. (Physiol.)

9. **Leon A. Heppel and Virginia T. Porterfield** (*introduced by* B. L. Horecker), *Laboratory of Physical Biology, National Institute of Health, Bethesda, Md.*

Enzymatic cleavage of organic halides. (Biochem.)

10. **F. Co Tui, J. H. Mulholland** (*by invitation*) **and N. Knox** (*by invitation*), *Department of Surgery, New York University College of Medicine*

Protein depletion syndrome and responses to hyperalimentation.

PHARMACOLOGY

Papers Read by Title

1. **Benedict E. Abreu and** (*by invitation*), **Grant W. Liddle, Arthur L. Burks, Alexander Simon, Violette Sutherland and Gilbert S. Gordon,** *Division of Pharmacology and Experimental Therapeutics, and Psychiatry, University of California Medical School, San Francisco*

Effects of amphetamine, dihydroergotamine, and methadon on human cerebral blood flow and oxygen uptake.

2. **Shannon C. Allen and Talbot G. Mortarotti** (*by invitation*), *Pharmacology Division, Bureau of Agricultural and Industrial Chemistry, U. S. Department of Agriculture, Albany, Calif.*

The effect of rutin on oxygen toxicity in rats.

3. **Anthony M. Ambrose,** *Pharmacology Division, Bureau of Agricultural and Industrial Chemistry, U. S. Department of Agriculture, Albany, Calif.*

The effect of rutin and quercetin on scorbutic guinea pigs.

4. **Hamilton H. Anderson, Herbert G. Johnstone**

(*by invitation*), **and A. Pena Chavarria** (*by invitation*), *University of California Medical School, San Francisco, and Hospital San Juan de Dios, San Jose de Costa Rica, A.C.*
Parasiticidal activity of thioarsenites in man.

5. **Arthur L. Bachelor and Henry W. Elliott** (*introduced by* Benedict E. Abreu), *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco*
The action of methadon upon the respiration of rat diaphragm and liver and kidney cortex slices.

6. **T. R. W. Barnes** (*introduced by* Charles H. Hine), *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco*
The toxicity of polyallyl alcohol.

7. **W. Barrett, A. Cameron, N. Hansen, E. Herrold, A. Mackenzie, B. Richards, F. Roth, J. Smith** (*all by invitation*), (*introduced by* F. F. Yonkman), *Division of Pharmacology, Research Department, Ciba Pharmaceutical Products, Inc., Summit, N. J.*
Toxicological and pharmacological data concerning 2-benzyl-imidazoline hydrochloride (Priscol).

8. **Harry Beckman,** *Marquette University School of Medicine*
Infectivity of sporozooites of *Plasmodium Cathemerium 3H2* exposed *in vitro* to hen and canary bloods.

9. **Harold F. Chase and Bani K. Bhattacharya** (*by invitation*), *Departments of Pharmacology and Surgery, Western Reserve University and The University Hospitals of Cleveland, Cleveland*
Prolongation of curare action with a peanut oil and beeswax vehicle.

10. **Yin-ch'ang Chin** (*introduced by* Hamilton H. Anderson), *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco*
Effect of some derivatives of subtilin on tubercle bacilli and rabbit leukocytes *in vitro*.

11. **F. Co Tui, V. Hollander, A. S. Keston, J. H. Mulholland,** *Department of Surgery, New York University College of Medicine*
Total body water by D_2O tracer study in protein depletion.

12. **John Emerson Davis,** *University of Arkansas School of Medicine, Little Rock*
Failure of anti-histaminic drugs to antagonise the anemia produced by fat plus choline.

13. **M. Edward Davis** (*by invitation*), **Nicholas W. Fugo, and Evelyn Ai-Feng Yu** (*by invitation*), *Department of Obstetrics and Gynecology and the Department of Pharmacology, University of Chicago and The Chicago Lying-in Hospital*
Some observations on the non-specific choline esterase in the human female.

14. **Victor A. Drill,** *Department of Pharmacology, Yale University School of Medicine, New Haven*
Relation of meat intoxication in Eck-fistula dogs to hepatic dysfunction.

15. **Frank G. Everett** (*introduced by* Norman David), *Department of Pharmacology, University of Oregon Medical School, Portland*
Comparison of depth of local anesthesia obtained with 2% and 4% procaine solutions.

16. **Chalmers L. Gemmill,** *Department of Pharmacology, University of Virginia, Medical School, Charlottesville*
Inhibitory effect of stilbamidine, guanidine and arginine on glycolysis.

17. **Alvin S. Hambly, Jr.** (*introduced by* Hamilton H. Anderson), *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco*
Chloroquine in human giardiasis.

18. **Carroll A. Handley and Marguerite LaForge** (*by invitation*), *Dept. of Pharmacology, Baylor University College of Medicine, Houston*
Some extra-cardiac effects of digitalis.

19. **Eder L. Hansen and Racheal K. Reed** (*introduced by* Hamilton H. Anderson), *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco*
Action of peroxides on endameba histolytica.

20. **Roy G. Herrmann** (*by invitation*) **and Kenneth P. DuBois,** *University of Chicago Toxicity Laboratory and the Department of Pharmacology, University of Chicago*
The action of p-dimethylaminobenzenediazo sodium sulfonate on carbohydrate metabolism.

21. **Charles H. Hine and Thomas Nathenial Burbridge** (*by invitation*), *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco*
The effects of 2,4 dinitrophenol and thiouracil on chronic methanol poisoning.

22. **Helen L. Holland** (*by invitation*) **and E. G. Gross,** *Department of Pharmacology, School of Medicine, Iowa City*
Two new analgesics.

23. **K. Hwang** (*by invitation*) **and K. R. Unna,** *Department of Pharmacology, University of Illinois College of Medicine, Chicago*
The effect of curare on the esophagus of the dog.

24. **Hal P. James and Ernest W. Page** (*introduced*

by C. H. Hine), *Divisions of Obstetrics and Gynecology and of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco*
Influence of some analgetics of obstetrical importance upon respiration of mature human placenta *in vitro*.

25. **Norman W. Karr** (*introduced by* Norman A. David), *Department of Pharmacology, University of Oregon Medical School, Portland*
Circulatory and adrenolytic actions of DHE and DHO.

26. **A. C. Kirchhof,** *The Division of Anesthesiology, University of Oregon Medical School, Portland*
Further studies on synthetic analgesics.

27. **C. D. Leake,** *University of Texas Medical Branch, Galveston*
Molecular architecture of cells.

28. **David Lehr,** *Department of Pharmacology, New York Medical College, Flower and Fifth Avenue Hospitals, New York*
Studies on the toxicity and pharmacology of some synthetic antispasmodics.

29. **Go Lu** (*Fellow, American Bureau for Medical Aid to China Inc.; by invitation*), *Department of Pharmacology and Therapeutics, Stanford University School of Medicine, San Francisco*
Comparative effects of sparteine and quinidine on isolated frog heart.

30. **David I. Macht and Thomas Hoffmaster** (*by invitation*), *Division of Pharmacology, Laboratories of Sinai Hospital, Baltimore.*
Influence of benadryl and pyribenzamine on the neuromuscular system of rats.

31. **W. A. McOmie** (*introduced by* Hamilton H. Anderson), *Division of Pharmacology and Experimental Therapeutics, University of California Medical School and College of Pharmacy, San Francisco*
Acute vapor toxicity to mice and local irritant effects in the rabbit of methyl pentadiene.

32. **Mark Nickerson,** *Department of Pharmacology, University of Utah School of Medicine, Salt Lake City*
Effect of procaine on cyclopropane-epinephrine cardiac arrhythmias.

33. **Carl C. Pfeiffer, Elizabeth H. Jenney** (*by invitation*) **and I. Gersh,** *Depts. of Pharmacology and Pathology, University of Illinois College of Medicine, Chicago*
Chronic intoxication of the CNS produced by the methadon sidechain.

34. **Frederick S. Philips, Maynard B. Chenoweth, and Carlton C. Hunt** (*by invitation*), *Departments of Pharmacology, Cornell Univ. Medical College and the Sloan-Kettering Institute for Cancer Research, New York*
Studies on the toxicology of podophyllotoxin and related substances.

35. **Raymond W. Pickering and Richard C. Burnett** (*introduced by* Benedict E. Abreu), *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco*
Spasmolytic and other pharmacologic properties of β-piperidinoethyl phenyl-α-thienyl-glycolate.

36. **Harry J. Pratt** (*introduced by* R. Beutner), *Department of Pharmacology, Hahnemann Medical College, Philadelphia*
Abolition of extra systoles in the perfused isolated rabbit heart by procaine and monocaine hydrochlorides.

37. **R. R. Rollins** (*introduced by* Hamilton H. Anderson), *Division of Pharmacology and Experimental Therapeutics, University of California Medical School, San Francisco*
Comparative intragastric and local toxicities of tert-butyl-hydrogen peroxide, di-tert-butyl-peroxide, and 2,2 bis-di-tert-butyl peroxybutane.

38. **Edward Saxey** (*by invitation*) **and Nilkanth M. Phatak,** *Department of Pharmacology, University of Oregon Medical School, Portland*
Use of oxygen consumption method for measuring antithyroid activity of n-propyl-thiouracil, thiouracil and 2-aminothiazole in rats.

39. **F. E. Shideman, A. R. Kelly** (*by invitation*), **and B. J. Adams** (*by invitation*), *Department of Pharmacology, University of Michigan, Ann Arbor*
Blood levels of thiopental (Pentothal) following repeated intravenous administration to the dog.

40. **Robert Tarail and W. Lane Williams** (*introduced by* R. N. Bieter), *Departments of Anatomy and Medicine, University of Minnesota*
Some effects of ouabain and/or calcium in mice.

41. **Ronald M. Thompson** (*by invitation*) **and Julius M. Coon,** *Dept of Pharmacology, University of Chicago*
Effect of adrenolytic agents on the response to pressor substances in the domestic fowl.

42. **Abraham Wikler,** *Research Department, U. S. Public Health Service Hospital, Lexington, Ky.*
Reactions of chronic decorticated dogs during a cycle of addiction to methadon.

43. **Robert H. Wilson, J. C. Lewis and E. M. Humphreys** (*by invitation*), *Pharmacology Division and the Western Regional Research Laboratory of the Bureau of Agriculture and*

Industrial Chemistry, U.S.D.A., Albany, Calif.

Subtilin in blood after parenteral administration.

44. **Martin M. Winbury and John Love** (*introduced by* W. E. Hambourger), *Lehigh Valley Laboratories, Easton, Pa.*

The effect of ANTU on the body temperature in the rat.

45. **S. Loewe,** *Department of Pharmacology, University of Utah School of Medicine, Salt Lake City*

Anticonvulsant actions of trimethadione-phenobarbital and trimethadione-diphenylhydantoin combinations.

46. **Alfred Leimdorfer,** *Department of Pharmacology, University of Illinois, College of Medicine, Chicago*

Influence of amidone, morphine and strychnine on the Straub-tail-test of white mice.

47. **Emmett B. Carmichael and Walter H. Johnson** (*by invitation*), *Biochemistry Department, Medical College of Alabama, Birmingham*

The LD_{50} of pentobarbital in nursed and unnursed newborn rats. (Biochem.)

48. **T. C. Barnes,** *Dept. of Pharmacology, Hahnemann Medical College and Hospital of Philadelphia*

Phase-boundary potentials of some analogs of prostigmine. (Physiol.)

49. **John V. Scudi, John F. Reinhard** (*by invitation*) **and N. B. Dreyer** (*by invitation*), *Nepera Chemical Co., Yonkers, and the Department of Pharmacology, University of Vermont*

Pharmacological characteristics of neohetramine, a new antihistaminic drug. (Biochem.)

THE AMERICAN SOCIETY FOR EXPERIMENTAL PATHOLOGY

THIRTY-THIRD ANNUAL MEETING

PATHOLOGY A

Tuesday, March 16, 9:00 a.m.

ROOM 15, CONVENTION HALL

Injury, Repair

1. **John W. Rebuck and Elizabeth A. Monaghan** (*introduced by* F. W. Hartman) *Department of Anatomy, University of Minnesota, Minneapolis, and Department Labs., Henry Ford Hospital, Detroit*

Peroxidase in the lymphocytes of man in acute inflammation.

2. **Edwin W. Schultz,** *Department of Bacteriology and Experimental Pathology, School of Medicine, Stanford University, Calif.*

Structure and repair of the olfactory mucosa in rhesus monkeys.

3. **Louise Pearce,** *Rockefeller Institute for Medical Research, Princeton*

Hereditary osteopetrosis of the rabbit.

4. **Robert S. Haukohl** (introduced by W. A. D. Anderson), *Department of Pathology and Bacteriology, Marquette University School of Medicine, Milwaukee*

The renal lesion of murine haemobartonellosis compared with lower nephron nephrosis.

5. **J. H. Grindlay, E. V. Flock, and J. L. Bollman,** *Division of Experimental Medicine, Mayo Foundation, Rochester*

Hepatic lymph and ascitic fluid following experimental chronic obstruction of the inferior vena cava.

6. **James H. Baxter** (introduced by Donald D. Van Slyke), *Hospital of the Rockefeller Inst. for Medical Research, New York*

Circulatory disturbances in hepatic and renal cortical necrosis.

7. **Arthur A. Nelson and Geoffrey Woodard** (by invitation), *Division of Pharmacology, Food and Drug Administration, Federal Security Agency, Washington*

Adrenal cortical atrophy and liver damage produced in dogs by feeding 2,2-BIS-(parachlorophenyl) - 1,1 - dichloroethane (DDD).

8. **Benjamin Highman,** *National Institute of Health, Pathology Laboratory, Washington*

Influence of various protective compounds in preventing fatty changes by dichloroethane.

9. **Hans Popper, Frederick Steigmann, Alvin Dubin** (*by invitation*) **and Hattie M. Dyniewicz** (*by invitation*), *Hektoen Institute for Medical Research of the Cook County Hospital, Chicago*

The influence of lipid ingestion on the thymol turbidity test.

10. **Rolf Lium** (*by invitation*) **and Stephen Maddock,**

Surgical Research Laboratory of the Boston City Hospital

Experimental production of acute pancreatitis.

11. **M. H. F. Friedman, B. F. Haskell** (*by invitation*), **and J. M. Waldron** (*by invitation*). *Department of Physiology, Jefferson Medical College and Department of Surgery, Jefferson Hospital, Philadelphia*

Treatment of non-specific ulcerative colitis for one year with intestinal extracts. (Physiol.)

PATHOLOGY B

Tuesday, March 16, 9:00 a.m.

Room 16, Convention Hall

Immunology

1. **Edward C. Rosenow**, *Rare Metals Institute of the California Institute of Technology, Pasadena, and the Longview Hospital, Cincinnati*

Production *in vitro* from protein and other solutions of substances resembling "natural" agglutinins and precipitins.

2. **Richard H. Follis, Jr.** *Department of Pathology, Johns Hopkins University*

Respiration of tissues from hypersensitive animals when antigen is added *in vitro*.

3. **L. W. Roth, R. K. Richards, and I. M. Shepperd** (*by invitation*) *Department of Pharmacology, Abbott Laboratories, North Chicago*

Production of anaphylaxis in guinea pigs with weakly antigenic protein hydrolysates. (Physiol.)

4. **Ernest Kun** (*Introduced by* C. Phillip Miller) *Department of Pharmacology and Medicine, University of Chicago, Chicago*

The effect of bacterial endotoxins on the carbohydrate metabolism of the rabbit.

5. **Walter D. Hawk** (*by invitation*) **and C. Phillip Miller**, *Department of Medicine, The University of Chicago, Chicago*

Protection against bacterial endotoxin by penicillin and its impurities.

6. **A. P. Krueger, T. Cohn** (*by invitation*) **and P. N. Smith** (*by invitation*), *Department of Bacteriology and Office of Naval Research Task V, University of California, Berkeley*

The production of phage in the absence of cellular growth.

7. **Kjell Agner** (*introduced by* A. M. Pappenheimer, Jr.) *Chemical Laboratory of the Serafimer Hospital, Stockholm, and the Department of Bacteriology, New York University College of Medicine*

Detoxification of diphtheria toxin by peroxidase. (Biochem.)

8. **Robert T. Thompson and Frances E. Moses** (*introduced by* M. A. Blankenhorn), *Departments of Internal Medicine and Biochemistry, College of Medicine, University of Cincinnati*

Specificity of human serum antihyaluronidase for antagonism of a particular species of bacterial hyaluronidase.

9. **William E. Ehrich, Carolyn Forman** (*by invitation*) **and Joseph Seifter**, *Philadelphia General Hospital, University of Pennsylvania, Wyeth Institute Applied Biochemistry*

A study of experimental serum sickness.

JOINT SESSION OF THE FEDERATION

Tuesday, March 16, 1:30 p.m.

Ballroom, Convention Hall

Program on page 314

PATHOLOGY BUSINESS MEETING

Tuesday, March 16, 4:15 p.m.

Room 15, Convention Hall

PATHOLOGY A

Wednesday, March 17, 9:00 a.m.

Room 20, Convention Hall

JOINT SESSION WITH THE AMERICAN PHYSIOLOGICAL SOCIETY

Coagulation

1. **R. H. K. Foster**, *Department of Pharmacology, St. Louis University School of Medicine*

Coagulation in plain and partially heparinized plasma.

2. **Arnold G. Ware** (*by invitation*) **and Walter H. Seegers**, *Department of Physiology, Wayne University, College of Medicine*

Serum accelerator globulin, quantitative determination, purification and properties.

3. **L. M. Tocantins, R. T. Carroll** (*by invitation*) **and Thomas McBride** (*by invitation*), *Division of Hematology, Department of Medicine, Jefferson Medical College, Philadelphia*

Lipid inhibitors and accelerators of blood coagulation in extracts of human brain.

4. **Bernard G. H. Thomas** (*by invitation*), **Jacqueline Spears** (*by invitation*) **and Charles R. Linegar,** *Pharmacological Development Division, E. R. Squibb and Sons, New Brunswick*
Factors affecting the use of lung thromboplastin in the determination of prothrombin time.
5. **John H. Ferguson** (*by invitation*) **and Jessica H. Lewis, P. W. Boyles, B. L. Travis and E. B. Gerheim,** *Department of Physiology, University of North Carolina, Chapel Hill*
Studies on the stability, inhibition and activation of fibrinolytic protease.
6. **Walters H. Seegers and Arnold G. Ware** (*by invitation*), *Department of Physiology, Wayne University, College of Medicine*
Purification of prothrombin and thrombin.
7. **F. L. Munro and Muriel Platt Munro** (*introduced by* F. R. Miller), *Charlotte Drake Cardeza Foundation, Department of Medicine, Jefferson Medical College and Hospital, Philadelphia*
Purification of a substance from bovine serum which reduces the prothrombin time of aged plasma.
8. **Mario Stefanini** (*by invitation*) **and Armand J. Quick,** *Department of Biochemistry, Marquette University School of Medicine, Milwaukee*
The concentration of the labile factor of prothrombin in the blood of various species. (Biochem.)
9. **Marjorie B. Zucker,** *Department of Physiology, College of Physicians and Surgeons, Columbia University*
The effect of thrombin injections on hemostasis.
10. **Armand J. Quick,** *Department of Biochemistry, Marquette University School of Medicine, Milwaukee*
Rôle of platelets in coagulation of blood, evidence of an inhibitor of the platelet factor. (Biochem.)
11. **F. D. Mann, Margaret Hurn and D. R. Mathieson** (*introduced by* T. B. Magath), *Division of Clinical Laboratories, Mayo Clinic, Rochester*
The platelets as foci in the coagulation of the blood.
12. **Alfred Lewin Copley,** *Marine Biological Laboratory, Woods Hole, and Laboratory of Cellular Physiology, Department of Biology, New York University, New York*
Embolization of platelet agglutination thrombi in the hamster's pouch produced by heparin.

PATHOLOGY B

Wednesday, March 17, 9:00 a.m.

ROOM 16, CONVENTION HALL

Vitamins, Hormones, Basement Membranes

1. **Charlotte L. Maddock** (*by invitation*), **S. B. Wolbach and Dorothy Jensen** (*by invitation*), *Department of Pathology, Harvard Medical School and Surgical Research Laboratory, Boston City Hospital*
Hypoprothrombinemia with hemorrhage as a cause of death in the rat in hypervitaminosis A.
2. **Herbert C. Stoerk** (*introduced by* A. M. Pappenheimer, Sr.), *Department of Bacteriology, Harvard Medical School, Boston*
Desoxypyridoxine-morphologic and functional changes in acute pyridoxine deficiency.
3. **James F. Rinehart and Louis D. Greenberg** (*by invitation*), *Division of Pathology, University of California, Medical School, San Francisco*
Arteriosclerotic lesions in pyridoxine-deficient monkeys.
4. **Janet M. Lemley, Robert G. Gale, Robert H. Furman, Mary E. Charrington** (*by invitation*) **and George R. Meneely,** *Departments of Medicine and Biochemistry, Vanderbilt University School of Medicine*
Plasma tocopherol levels in cardiac patients.
5. **Hans Kaunitz and Charles A. Slanetz** (*by invitation*), *Departments of Pathology and Animal Care, College of Physicians and Surgeons, Columbia University, New York City*
Failure of implantation in vitamin E deficient rats.
6. **William Trager,** *Department of Animal and Plant Pathology, The Rockefeller Institute for Medical Research, Princeton*
Further studies on a fat-soluble biotin-active material (FSF) from plasma.
7. **Albert Segaloff and Richard L. Coppedge** (*by invitation*), *Departments of Medicine and Physiology, Tulane University and the Alton Ochsner Medical Foundation, New Orleans*
Further studies on the hepatic potentiation of the estrogenic activity of triphenylchloroethylene.
8. **Isidore Gersh,** *Department of Pathology, University of Illinois College of Medicine, Chicago*
Histochemical studies of basement membranes.
9. **J. F. A. McManus** (*introduced by* R. D. Baker), *Department of Pathology, The Medical College of Alabama, Birmingham*
Histochemical features of the renal basement membrane.

PATHOLOGY A

Wednesday, March 17, 1:45 p.m.

ROOM 15, CONVENTION HALL

Blood

1. **J. M. Waldron** (*by invitation*) **and M. H. F. Friedman**, *Department of Physiology, Jefferson Medical College, Philadelphia*
   The relationship between anticoagulants and lipemia. (Physiol.)
2. **E. B. Gerheim,** (*by invitation*), **J. H. Ferguson and B. L. Travis** (*by invitation*), *Department of Physiology,,University of North Carolina, Chapel Hill*
   Staphylcoagulase and staphylokinase. (Physiol.)
3. **Virginia Minnich** (*by invitation*) **and Carl V. Moore**, *Department of Internal Medicine, Washington University School of Medicine, St. Louis*
   Hypoplastic anemia induced in guinea pigs by 4-amino pteroyl glutamic acid.
4. **Reubenia Dubach** (*by invitation*), **Sheila Callender** (*by invitation*) **and Carl V. Moore,** *Department of Internal Medicine, Washington University School of Medicine, St. Louis*
   Iron absorption in normal subjects and in patients with anemias of varied etiology.
5. **F. S. Robscheit-Robbins,** *Department of Pathology, School of Medicine and Dentistry, University of Rochester, Rochester*
   The influence of folic acid on hemorrhage anemia in dogs.
6. **Valy Menkin,** *Agnes Barr Chase Foundation for Cancer Research, Temple University School of Medicine, Philadelphia*
   The fate of aged leukocytosis-promoting factor of exudates.
7. **Harry Sobel** (*by invitation*) **and Jacob Furth,** *Cornell University Medical College, New York City and Veterans Administration Hospital, Southwestern Medical College, Dallas*
   Experimental hypervolemia, time of onset and some associated physiological and chemical changes.
8. **Earl P. Benditt,** *Department of Pathology, The University of Chicago Clinics, Chicago*
   The relationship of caloric intake level and protein intake level to rate of protein synthesis.
9. **Roger Terry,** (*introduced by* G. H. Whipple), *Department of Pathology, School of Medicine and Dentistry, University of Rochester, Rochester*
   Prolonged parenteral plasma produces hyperproteinemia and proteinuria in dogs and maintains nitrogen equilibrium and health.

PATHOLOGY B

Wednesday, March 17, 1:45 p.m.

ROOM 16, CONVENTION HALL

Infection

1. **Edmund P. Finch** (*by invitation*) **and Seward E. Owen**, *Veterans Administration, Hines, Ill.*
   Urinary tract infections and antibiotics. (Physiol.)
2. **Clayton G. Loosli,** *Department of Medicine, University of Chicago, Chicago*
   The histogenesis of cells in pneumonia as seen in multiple lobe pneumococcus (type 1) infections in dogs.
3. **Joseph E. Smadel and Elizabeth B. Jackson** (*by invitation*), *Department of Virus and Rickettsial Disease, Army Medical Department Research and Graduate School, Army Medical Center, Washington*
   Chemotherapeutic effect of chloromycetin on experimental infection with psittacosis and lymphogranuloma venereum viruses.
4. **F. B. Gordon, F. M. Schabel, Jr.,** (*by invitation*) **and Margaret Abendroth** (*by invitation*), *Department of Bacteriology and Parasitology, University of Chicago*
   Susceptibility of *macacus cynomolgus* to Japanese encephalitis virus with special reference to the alimentary canal.
5. **O. M. Gruhzit and R. A. Fisken** (*by invitation*), *Research Laboratories, Parke, Davis and Co., Detroit*
   Failure of trypanosoma cruzi lysate in treatment of Brown-Pierce carcinoma of rabbits.
6. **Orlyn Wood** (*by invitation*), **Doris Noshold** *by invitation*) **and Lloyd D. Seager,** *Department of Pharmacology, Woman's Medical College of Pennsylvania*
   The effect of splenectomy upon the susceptibility of mice to infection by trypanosoma cruzi. (Pharm.)
7. **Alfred Golden and R. R. Overman** (*by invitation*), *Departments of Pathology and Physiology, University of Tennessee, College of Medicine*
   Sodium-potassium levels and adrenal gland necrosis in experimental Simian malaria.
8. **R. H. Rigdon,** *Department of Pathology, School of Medicine, University of Texas*
   Effect of blood and oxygen on P. knowlesi infection in monkeys.

9. **Milton D. Levine, Ray F. Garzoli, Robert E. Kuntz and John H. Killough,** (*introduced by* K. M. Endicott), *Naval Medical Research Institute, National Naval Medical Center*
On the demonstration of hyaluronidase in cercariae of schistosoma mansoni.

Federation "Mixer"

Wednesday, March 17, 9:00 p.m.

ARENA, CONVENTION HALL

PATHOLOGY A

Thursday, March 18, 9:00 a.m.

ROOM 15, CONVENTION HALL

Cholesterol, Isotopes, Miscellaneous

1. **Jesse L. Bollman and Eunice V. Flock,** *Division of Experimental Medicine, Mayo Foundation, Rochester, Minnesota*
Cholesterol in Intestinal and Hepatic Lymph of the Rat.
2. **Forrest E. Kendall, Walter Meyer, and Margaret Bevans** (*introduced by* H. P. Smith), *Research Service, First (Columbia) Division, Goldwater Mem. Hospital, Dept. of Hospitals, City of New York and the Dept. of Med., College of Physicians and Surgeons, Columbia University*
Effect of intravenous injection of oxidized cholesterol upon the production of atherosclerosis in rabbits.
3. **Margaret Bevans, Liese Lewis Abell, and Forrest E. Kendall** (*introduced by* H. P. Smith), *From the Research Service, First (Columbia) Division, Goldwater Mem. Hospital, Dept. of Hospitals, City of New York, and the Dept. of Med., College of Physicians and Surgeons, Columbia University*
Production of intimal atherosclerosis by intravenous injection of colloidal cholesterol into rabbits.
4. **Alfred Steiner, Margaret Bevans, and Forrest E. Kendall,** (*introduced by* H. P. Smith), *From the Research Service, First (Columbia) Division, Goldwater Mem. Hospital, Dept. of Hospitals City of New York, and the Dept. of Medicine, College of Physicians and Surgeons, Columbia University*
Production of arteriosclerosis in dogs with cholesterol and thiouracil.
5. **Russell L. Holman,** *Dept. of Pathology, L. S. U. School of Medicine, New Orleans*
Arterial disease may be a matter of days, not decades.
6. **P. F. Hahn,** *Vanderbilt Univ. School of Med., Nashville, Tenn.*
Tumor therapy by the direct infiltration of radioactive colloidal metallic gold.
7. **George Rouser** (*introduced by* P. F. Hahn), *Vanderbilt Univ. School of Med., Nashville*
Preparation of radioactive gold colloids for use in the therapy of malignancies.
8. **Herman N. Eisen** (*by invitation*) **and Albert S. Keston** (*by invitation*), **Robert C. Warner,** *Depts. of Medicine and Chemistry, New York University, College of Med.*
Immunochemical studies with proteins labelled with trace amounts of radioactive iodine. (Biochem.)
9. **H. G. Davis, Jr.** (*by invitation*) **and R. D. Baker,** *Dept. of Pathology, Medical College of Alabama*
Blood iodine in alloxan diabetes of dogs.
10. **Louis J. Strobino** (*introduced by* Lee E. Farr) *Alfred I. duPont Institute of the Nemours Foundation, Wilmington, Delaware*
Some factors associated with variations in nitrogen and ash content of bone.
11. **Frederick M. Allen,** *City Hospital, Welfare Island, New York, N. Y.*
Discrepant animal and clinical observations on hypothermia and procaine.
12. **Frank Dixon** (*by invitation*) **and Shields Warren,** *Laboratory of Pathology, New England Deaconess Hospital, Boston*
Antigen tracer studies in anaphylactic shock.

PATHOLOGY

Motion Pictures

Thursday, March 18, 8:00 p.m.

ROOM C, CONVENTION HALL

Program on page 314

Pathology

Papers Read by Title

1. **Julius Schultz** (*by invitation*) **and Charles Weiss,** *Laboratories of the Jewish Hospital, Philadelphia 41, Pa.*
Hydrolosis of Dl-Leucylglycylglycine by sera of tuberculous and normal rabbits.
2. **Alfred Golden,** *Department of Pathology, University of Tennessee, College of Medicine, Memphis, Tennessee*
Adrenal gland lesions in experimental simian malaria and similar human lesions in varied diseases.
3. **William Trager,** *Dept. of Animal and Plant Pathology, Rockefeller Institute for Medical Research, Princeton*
The inhibition of growth of lactobacillus casei by lyolecithin.

AMERICAN INSTITUTE OF NUTRITION

Twelfth Annual Meeting

NUTRITION

Tuesday, March 16, 9:00 a.m.

Room A, Convention Hall

Vitamins

1. **Paul F. Fenton** (*by invitation*) **and George R. Cowgill,** *Yale Nutrition Laboratory, Department of Physiological Chemistry, Yale University, New Haven*
   The pantothenate requirements of the mouse, with observations on the role of biotin, inositol and p-aminobenzoic acid.
2. **Elaine P. Ralli, Mary E. Dumm** (*by invitation*), **and Paul Roth** (*by invitation*), *Department of Medicine, New York University College of Medicine, New York*
   Pantothenic acid requirement of adrenalectomized rats and its relation to the white blood cells.
3. **Thomas R. Riggs** (*by invitation*) **and D. Mark Hegsted,** *Department of Nutrition, Harvard School of Public Health and Department of Biological Chemistry, Harvard Medical School, Boston*
   The effect of pantothenic acid deficiency on acetylation in rats.
4. **Walter C. Russel and Arthur E. Teeri** (*by invitation*) (*now at the University of New Hampshire, Durham*), *Department of Agricultural Experiment Station, Rutgers University, New Brunswick*
   Blood constituents of swine in a pantothenic acid deficient condition.
5. **J. B. Neilands** (*by invitation*) **and F. M. Strong,** *Department of Biochemistry, University of Wisconsin, Madison*
   The enzymatic liberation of pantothenic acid. (Biochem.)
6. **W. T. Burnett** (*by invitation*), **R. C. Miller, and R. A. Dutcher,** *Pennsylvania State College, State College*
   Niacin and protein relationships in corn diets for growing pigs.
7. **W. D. Salmon,** *Laboratory of Animal Nutrition, Alabama Polytechnic Institute, Auburn*
   The tryptophane requirement for growth of the rat.
8. **Herbert P. Sarett and Grace A. Goldsmith,** *Nutrition Research Laboratory, Tulane University School of Medicine, New Orleans*
   Effects of gelatin, glycine, and pyridoxine on tryptophane and nicotinic acid metabolism in humans.
9. **Ernestine I. Frazier** (*by invitation*), **Thelma Porter, and Mary Jane Humphrey** (*by invitation*), *Department of Home Economics, University of Chicago*
   The utilization of nicotinic acid by pregnant women.
10. **Fred Rosen** (*by invitation*), **W. A. Perlzweig, and Philip Handler,** *Department of Biochemistry, Duke University School of Medicine, Durham*
    A fluorimetric assay for N^1-methyl-6-pyridone-3-carboxylamide. (Biochem.)
11. **R. W. Engel** (*introduced by* W. D. Salmon), *Laboratory of Animal Nutrition, Alabama Polytechnic Institute, Auburn*
    Anemia in chronic choline deficiency in the rat.

Joint Session of the Federation

Tuesday, March 16, 1:30 p.m.

Ballroom, Convention Hall

Program on p. 314

American Institute of Nutrition Dinner; Presentation of Awards; Business Meeting

Tuesday, March 16, 6:30 p.m.

NUTRITION

Wednesday, March 17, 9:00 a.m.

Room A, Convention Hall

Vitamins

1. **R. M. Johnson** (*by invitation*) **and C. A. Baumann,** *Department of Biochemistry, College of Agriculture, University of Wisconsin, Madison*
   Relative importance of growth and metabolic rate on the utilization of vitamin A by the rat.
2. **Barbara Kelley** (*by invitation*) **and Harry G. Day,** *Department of Chemistry, Indiana University, Bloomington*
   Effect of xanthophyll on the utilization of carotene and vitamin A by the rat.
3. **Jean Mayer** (*by invitation*) **and W. A. Krehl,** *Yale Nutrition Laboratory, Department of Physiological Chemistry, Yale University, New Haven*
   Vitamin A deficiency and diet composition. (Biochem.)

4. **Albert E. Sobel, Lottie Besman** (*by invitation*), **and Benjamin Kramer,** *Departments of Biochemistry and Pediatrics, The Jewish Hospital of Brooklyn, New York*
Vitamin A absorption in newborns, older children, adults, and A storage in rats. (Biochem.)
5. **E. L. Hove,** *Research Laboratories, Distillation Products, Inc., Rochester, New York*
α-Tocopherol and certain nitrogenous compounds as factors influencing the mortality of rats after carbon tetrachloride poisoning.
6. **M. L. Quaife** (*introduced by* P. L. Harris), *Research Laboratories, Distillation Products, Inc., Rochester, New York*
Analytical procedures for determining individual tocopherols in a mixture of α-, β-, γ-, and δ-tocopherols. (Biochem.)
7. **Mary B. Mills** (*by invitation*), **Charlotte M. Damron** (*by invitation*), **and Joseph H. Roe,** *Department of Biochemistry, George Washington University School of Medicine, Washington*
Studies on the occurrence of diketo-L-gulonic acid dehydro-L-ascorbic acid and L-ascorbic acid.
8. **Katherine J. Elliott** (*by invitation*), **Shih Dzung Chen** (*by invitation*), **and Cecilia Schuck,** *Department of Home Economics, Purdue University, Lafayette*
Changes in dehydro and total ascorbic acid of cantaloupes on standing.
9. **J. Walter Wilson and Elizabeth H. Leduc** (*introduced by* P. H. Mitchell), *Brown University, Providence*
The effect of biotin on mitotic activity in the mouse liver. (Biochem.)
10. **Frank Davis** (*by invitation*) **and George M. Briggs,** *Department of Poultry Husbandry, University of Maryland, College Park*
Studies on the cellulose growth factor for chicks.
11. **H. R. Bird, A. C. Groschke** (*by invitation*), **and Max Rubin** (*by invitation*), *United States Department of Agriculture, Bureau of Animal Industry, Washington*
Effects of arsonic acid derivatives in stimulating growth of chicks fed certain diets.

NUTRITION

Wednesday, March 17, 1:45 p.m.

ROOM A, CONVENTION HALL

Nutrition and Disease; Lipids

1. **Howard A. Schneider,** *The Rockefeller Institute for Medical Research, New York*
The double strain phenomenon: Biological basis of the nutritional effect in natural resistance to inflection.
2. **Philip Handler and F. Bernheim** (*by invitation*), *Departments of Biochemistry and of Physiology and Pharmacology, Duke University School of Medicine, Durham*
Dietary factors in experimental renal hypertension. I. Protein.
3. **A. T. Milhorat,** *Departments of Psychiatry and Medicine, Cornell University Medical College, The Russel Sage Institute of Pathology and the New York Hospital, New York*
Effect of arginine on urinary output of 17-ketosteroids in a patient with myotonia atrophica.
4. **George V. Mann** (*by invitation*) **and Frederick J. Stare,** *Department of Nutrition, Harvard School of Public Health, and Department of Biological Chemistry, Harvard Medical School, Boston*
Diet and dose-response of weanling rats to intravenous alloxan.
5. **Julia O. Holmes, L. R. Parkinson** (*by invitation*), **Anne W. Wertz** (*by invitation*), **and Lois Brow** (*by invitation*), *Agricultural Experiment Station, University of Massachusetts, Amherst*
Dental caries in the rat, Mus Norvegicus.
6. **J. Knox Smith** (*by invitation*), **E. Potts Anderson** (*by invitation*), **Marie Zepplin** (*by invitation*), **C. A. Elvehjem, and Paul H. Phillips,** *Department of Biochemistry, University of Wisconsin, Madison*
Physical factors influencing dental caries in the cotton rat.
7. **Arild E. Hansen, Oleta Beck** (*by invitation*), **and Hilda Wiese** (*by invitation*), *Department of Pediatrics, University of Texas Medical School, Galveston*
Susceptibility to infection manifested by dogs on a low fat diet.
8. **Hilda F. Wiese** (*by invitation*) **and Arild E. Hansen,** *Department of Pediatrics, University of Texas Medical School, Galveston*
Lipid components of skin of dogs on low fat diet and dogs receiving lard.
9. **Robert P. Geyer** (*by invitation*), **George V. Mann** (*by invitation*) **and Frederick J. Stare,** *Department of Nutrition, Harvard School of Public Health and Department of Biological Chemistry, Harvard Medical School, Boston*
The turbidimetric determination of infused fat in blood after intravenous administration of fat emulsions.
10. **C. Boyd Shaffer and Frances H. Critchfield** (*introduced by* Paul L. McLain), *Chemical Hygiene Fellowship, Mellon Institute, Pittsburgh*
The nutritive value of the fatty acids of lard esterified with a polyethylene glycol. (Pharm.)

11. **Harry H. LeVeen** (*introduced by* Frank Co Tui), *Department of Surgery, New York University College of Medicine, New York*
The use of natural and synthetic fat emulsions for intravenous feeding. (Pharm.)

Federation "Mixer"

Wednesday, March 17, 9:00 p.m.

ARENA, CONVENTION HALL

NUTRITION

Thursday, March 18, 9:00 a.m.

ROOM A, CONVENTION HALL

General

1. **B. L. O'Dell** (*by invitation*) **and A. G. Hogan** *Department of Agricultural Chemistry, University of Missouri, Columbia*
Nutrients required by the rat for reproduction and lactation.
2. **Sister Ann Miriam Allegeier, S.C.N.** (*by invitation*), **Albert J. Sica** (*by invitation*), **Leonora Mirone** (*by invitation*), **Frank P. Panzarella** (*by invitation*), **and Leopold R. Cerecedo,** *Department of Biochemistry, Fordham University, New York*
Studies on reproduction and lactation in rats and mice maintained on synthetic diets.
3. **W. A. Krehl and I. D. Welt** (*by invitation*), *Yale Nutrition Laboratry, Department of Physiological Chemistry, Yale University, New Haven*
Nutritional studies on the cat. (Biochem.)
4. **Pearl Swanson, W. W. Smith** (*by invitation*), **M. Brush** (*by invitation*), **and H. Merrian** (*by invitation*), *The Nutrition Laboratory of the Iowa Agricultural Experiment Station, Iowa State College, Ames*
Evaluation of adequate protein nutrition.
5. **Mildred L. Bucker** (*by invitation*), **Ruth Shively** (*by invitation*), **and Janice M. Smith,** *Agricultural Experiment Station, Department of Home Economics, University of Illinois, Urbana*
Protein requirements of ten college women and adequacy of estimated quantities for ten weeks.
6. **L. R. Richardson,** *Department of Biochemistry and Nutrition, A and M College of Texas, College Station*
Southern peas as a source of protein for growth.
7. **Donald D. Kozoll** (*by invitation*), **William S. Hoffman, Gordon McNeil** (*by invitation*), and **Hans Popper** (*by invitation*), *Hektoen Institute for Medical Research of the Cook County Hospital, Chicago*
The determination of the biological indices of gluten and gluten fortified with lysine in normal and protein-deficient patients. (Biochem.)
8. **K. A. Kuiken** (*by invitation*), **and Carl M. Lyman,** *Department of Biochemistry and Nutrition, A and M College of Texas, College Station*
The availability of amino acids in some foods. (Biochem.)
9. **Hazel E. Munsell, Robert S. Harris, and Louis O. Williams** (*by invitation*), *Nutritional Biochemistry Laboratories, Massachusetts Institute of Technology, Cambridge*
Composition of Central American foods. I. Honduras.
10. **Barnett Sure,** *University of Arkansas, Fayetteville*
A new dairy food.
11. **A. L. Franklin** (*by invitation*), **J. W. Boehne III** (*by invitation*), **and T. H. Jukes,** *Lederle Laboratories Division, American Cyanamid Company, Pearl River, New York*
Thiouracil content of poultry tissue following prolonged feeding of the compound.

NUTRITION

Thursday, March 18, 1:45 p.m.

ROOM A, CONVENTION HALL

Mineral Nutrition

1. **Ruth Frenchman, Euphemia D. Boroughs, and Frances A. Johnston** (*introduced by* Hazel M. Hauck), *New York State College of Home Economics, Cornell University, Ithaca*
The absorption of iron from beef by women.
2. **Frances A. Johnston, Euphemia D. Boroughs, and Ruth Frenchman** (*introduced by* Hazel M. Hauch), *New York State College of Home Economics, Cornell University, Ithaca*
The adequacy of an intake of seven milligrams of iron per day for women.
3. **D. Mark Hegsted, Clement A. Finch** (*by invitation*), **and Thomas D. Kinney** (*by invitation*), *Department of Nutrition, Harvard School of Public Health; the Departments of Biological Chemistry, Medicine and Pathology, Harvard Medical School;*

and the Medical Clinic and Department of Pathology, Peter Bent Brigham Hospital, Boston

The effect of diet upon iron absorption.

4. **Leon M. Sharpe** (*by invitation*), **Robert S. Harris, Wendell C. Peacock** (*by invitation*), **and Richard C. Cooke** (*by invitation*), *Departments of Food Technology and Physics, The Massachusetts Institute of Technology and the Walter E. Fernald State School, Cambridge*

Effect of phytate and other food ingredients on the absorption of radioactive iron.

5. **L. B. Pett and G. F. Ogilvie** (*by invitation*), *Nutrition Division, Department of National Health and Welfare, Ottawa, Canada*

Hemoglobin levels by age and sex.

6. **Beula V. McKey** (*by invitation*), **Eleanor Smith** (*by invitation*), **and Janice M. Smith,** *Agricultural Experiment Station, Department of Home Economics, University of Illinois, Urbana*

Calcium requirements of seven adolescent girls.

7. **Margaret G. Morehouse** (*by invitation*), **Amber Lieng-shan Cheng** (*by invitation*) **and Harry J. Devel, Jr.,** *University of Southern California School of Medicine, Los Angeles*

Studies on fat digestibility; the effect of melting points and dietary calcium and magnesium levels.

8. **H. O. Kunkel** (*by invitation*) **and P. B. Pearson,** *Department of Biochemistry and Nutrition, A and M College of Texas, College Station*

The quantitative requirement of the rat for magnesium and effects of magnesium deficiency in the rabbit.

9. **Robert M. Hill and Dorsey E. Holtkamp** (*by invitation*), *Department of Biochemistry, University of Colorado Medical Center, Denver*

Manganese and thiamine in the diet of mother rats and body temperature control in the young. (Biochem.)

10. **Edith M. Carlisle** (*by invitation*) and **Helen T. Parsons,** *Department of Home Economics, University of Wisconsin, Madison*

Achromotrichia produced in mice on a cooked egg diet.

11. **Esther DaCosta and Ruth Clayton** (*introduced by* R. E. Johnson), *U. S. Army Medical Nutrition Laboratory, 1849 W. Pershing Road, Chicago*

Changes in the water content of the albino rat during the first week of dietary rehabilitation.

NUTRITION BUSINESS MEETING

Thursday, March 18, 4:30 p.m.

ROOM A, CONVENTION HALL

NUTRITION

Friday, March 19, 9:00 a.m.

ROOM A, CONVENTION HALL

Vitamins

1. **Grace A. Goldsmith and Herbert P. Sarett,** *Nutrition Research Laboratory, Tulane University School of Medicine, New Orleans*

Urinary excretion of B vitamins in persons on normal and restricted diets.

2. **Wanda I. Lameck** (*by invitation*), **Margaret N. Coryell** (*by invitation*), **Eliot F. Beach, and Icie G. Macy,** *Research Laboratory, Children's Fund of Michigan, Detroit*

Urinary thiamine and riboflavin excretion of children during fasting and under conditions of loading test.

3. **W. O. Caster** (*by invitation*), **Olaf Mickelsen, and Ancel Keys,** *Laboratory of Physiological Hygiene, University of Minnesota, Minneapolis*

Influence of dietary and physical factors on the urinary excretion of thiamine and pyramin. (Biochem.)

4. **Gladys Everson** (*by invitation*), **Eleanor Williams** (*by invitation*), **Elisabeth Wheeler** (*by invitation*), **Pearl Swanson, Margaret Eppright** (*by invitation*), **and Mattie Spivey** (*by invitation*), *The Nutrition Laboratory, Iowa Agricultural Experiment Station, Iowa State College, Ames*

Occurrence of B vitamins in tissues of rats fed rations satisfactory and unsatisfactory for reproduction.

5. **H. B. Pierce, R. F. Krause** (*by invitation*), **J. H. Browe** (*by invitation*), **and Susan Merrow** (*by invitation*), *Departments of Biochemistry, Medicine, and Anatomy, College of Medicine University of Vermont, Burlington*

The effect of vitamin therapy on serum blood levels.

6. **Harold P. Morris, Celia S. Dubnik** (*by invitation*), **and Thelma B. Dunn** (*by invitation*), *National Cancer Institute, USPHS, Bethesda*

Studies of thiamine deficiency in C3H mice.

7. **J. R. Haag and P. H. Weswig** (*by invitation*), *Oregon Agricultural Experiment Station, Corvallis*

Further observations concerning antithiamine activity in plants (Biochem.)

8. **Leo T. Samuels, A. A. Anderson** (*by invitation*), **E. Chapman** (*by invitation*), **and D. MacKay** (*by invitation*), *Department of Biochemistry, University of Utah School of Medicine, Salt Lake City*
   The effects of thiamine deficiency in rats force-fed to maintain caloric balance. (Biochem.)
9. **L. R. Richardson, R. M. Sherwood** (*by invitation*, **and H. L. German** (*by invitation*), *Departments of Biochemistry and Nutrition and Poultry Husbandry, A and M College of Texas, College Station*
   The riboflavin content of the diet and leg trouble in broad breasted bronze turkeys.
10. **Selma E. Snyderman** (*by invitation*), **Katherind D. Ketron** (*by invitation*), **Anthony A. Albanese, and L. Emmett Holt, Jr.**, *Department of Pediatrics, New York University and The Children's Medical Service of Bellevue Hospital, New York*
    The riboflavin requirement of the normal infant. (Biochem.)
11. **Elmer DeRitter** (*by invitation*), **Mary E. Moore** (*by invitation*), **Erich Hirschberg** (*by invitation*), **and Saul R. Rubin**, *Nutrition Laboratories, Hoffmann-LaRoche, Inc., Nutley, New Jersey*
    Critique of methods for determination of riboflavin in urine. (Biochem.)

NUTRITION

Papers Read by Title

1. **Marianne Goettsch**, *Department of Chemistry, School of Tropical Medicine, San Juan, Puerto Rico*
   Minimal protein requirement for growth in the rat.
2. **Marinus C. Kik**, *Department of Agricultural Chemistry, University of Arkansas, Fayetteville*
   Biological analysis of rice.
3. **Carl A. Kuether** (*by invitation*), **and Victor Myers**, *Department of Biochemistry, Western Reserve University, Cleveland*
   The nutritive value of cereal proteins in human subjects.
4. **J. F. McClendon**, *Hahnemann Medical College, Philadelphia*
   Food fermentation as a preventive of furunculosis.
5. **Leonora Mirone** (*by invitation*), **and Leopold R. Cerecedo**, *Department of Biochemistry, Fordham University, New York*
   Inadequacy of thymine as a nutritional factor for reproduction and lactation in the mouse.
6. **Barnett Sure and Frances House** (*by invitation*), *University of Arkansas, Fayetteville*
   Biological value of proteins in food yeasts.

THE AMERICAN ASSOCIATION OF IMMUNOLOGISTS

THIRTY-SECOND ANNUAL MEETING

IMMUNOLOGY

Tuesday, March 16, 9:00 a.m.

ROOM 10, CONVENTION HALL

1. **Lloyd D. Felton**, *National Institute of Health, Bethesda, Md.*
   PRESIDENTIAL ADDRESS: The significance of antigen to animal tissues.
2. **F. K. Sanders** (*introduced by* Joseph E. Smadel), *Dept. of Virus and Rickettsial Diseases, Army Medical Dept. Research and Graduate School, Army Medical Center, Washington*
   A poliomyelitis-like agent in hamsters inoculated with Lansing virus.
3. **Joel Warren** (*by invitation*) **and Joseph E. Smadel**, *Dept. of Virus and Rickettsial Diseases, Army Medical Dept. Research and Graduate School, Army Medical Center*
   A serological relationship between the virus of encephalomyocarditis and certain strains of poliomyelitis-like viruses.
4. **Joseph L. Melnick and Robert Ward** (*introduced by* Lloyd E. Felton), *Section of Preventive Medicine, Yale University School of Medicine*
   Adaptation of poliomyelitis strains to rodents with a word on nomenclature.
5. **A. P. McKee** (*by invitation*) **and William M. Hale**, *Dept. of Bacteriology, Univ. of Iowa*
   Reactivation of overneutralized influenza virus in chicken embryos.
6. **Morton Klein, J. H. Brewer, J. E. Perez and Beatrice Day** (*introduced by* Stuart Mudd), *Dept. of Bacteriology, University of Pennsylvania School of Medicine, and Hynson, Westcott and Dunning, Baltimore*

The inactivation of influenza virus by mercurials and reactivation by sodium thioglycolate and BAL.

7. **Morton Klein and Enrique J. Perez** (*introduced by* Stuart Mudd), *Dept. of Bacteriology, University of Pennsylvania School of Medicine*
   The reactivation *in vivo* of influenza A virus by BAL.
8. **B. A. Briody** (*introduced by* Thomas Francis, Jr.), *Walter and Eliza Hall Institute, Melbourne, Australia*
   Characterization of the "enzymic" action of influenza viruses on human red cells.
9. **Francis C. Lowell and Marguerite Buckingham** (*by invitation*), *Evans Memorial Hospital, Boston*
   Effect of salt concentration on the agglutination of red cells by influenza virus and antibody.
10. **Monroe D. Eaton,** *Dept. of Bacteriology and Immunology, Harvard Medical School*
    Studies on inhibition of the agents of murine and feline pneumonitis by penicillin.

JOINT SESSION OF THE FEDERATION

Tuesday, March 16, 1:30 p.m.

BALLROOM, CONVENTION HALL

Program on page 314

IMMUNOLOGY BUSINESS MEETING

Tuesday, March 16, 4:15 p.m.

ROOM 10, CONVENTION HALL

IMMUNOLOGY

Tuesday, March 16, 7:00 p.m.

ROOM 21, CONVENTION HALL

Symposia

I. WHAT WE KNOW ABOUT COMPLEMENT—AND WHAT DO WE WANT TO DO ABOUT IT?

1. **Michael Heidelberger,** LEADER, *College of Physicians and Surgeons, New York*
2. **Manfred M. Mayer,** *Johns Hopkins University School of Hygiene and Public Health*
3. **E. E. Ecker,** *Western Reserve University Medical School, Cleveland*
4. **Louis Pillemer,** *Institute of Pathology, Western Reserve University, Cleveland*

II. STATUS OF OUR KNOWLEDGE OF ALLERGY

1. **Mary H. Loveless,** LEADER, *Cornell Medical School and New York Hospital*
2. **Dan H. Campbell,** *California Institute of Technology, Pasadena*
3. **Frances C. Lowell,** *City Hospital, Boston*

IMMUNOLOGY

Wednesday, March 17, 9:00 a.m.

ROOM 10, CONVENTION HALL

1. **Alfred S. Evans** (*by invitation*) **and Edward C. Curnen,** *Section of Preventive Medicine, Yale University School of Medicine*
   Serological studies on infectious mononucleosis and other conditions with human erythrocytes modified by Newcastle disease virus.
2. **Thomas E. Burney** (*by invitation*) **and Orville J. Golub,** *Camp Detrick*
   The effect of certain enzyme inhibitors on the activity and growth of psittacosis virus.
3. **Charles A. Bailey, Fred H. Dierks and James E. Proffitt** (*introduced by* A. F. Coca), *National Naval Medical Center, Naval Medical Research Institute, Bethesda, Md.*
   On the purification of the rickettsia of tsutsugamushi disease (scrub typhus).
4. **Koda Guo** (*by invitation*), **Anna Dean Dulaney and Henry Packer** (*by invitation*), *Divisions of Pathology and Bacteriology, and Preventive Medicine, University of Tennessee College of Medicine*
   Studies on *Donovania granulomatis*. I. Locke-embryonic yolk medium for cultivation of the organism.
5. **Anna Dean Dulaney, Koda Guo** (*by invitation*) **and Henry Packer** (*by invitation*), *Divisions of Pathology and Bacteriology, and Preventive Medicine, University of Tennessee College of Medicine*
   Studies on *Donovania granulomatis*. II. Preparation of culture antigens and immunologic tests.
6. **W. D. Sutliff** (*by invitation*), **Anna Dean Dulaney and W. L. Davis** (*by invitation*), *Kennedy Hospital and Division of Pathology and Bacteriology, University of Tennessee*
   Complement-fixation with *P. knowlesi* antigen for diagnosis of malaria.
7. **Bernard D. Davis** (*introduced by* Jules Freund), *Public Health Research Institute of the City of New York*
   Complement fixation with soluble antigens of *Plasmodium knowlesi* and *Plasmodium lophurae*.
8. **W. H. Taliaferro, L. G. Taliaferro** (*by invitation*) **and F. E. Kelsey** (*by invitation*), *University of Chicago*
   Rôle of the spleen and phagocytosis in the quinine treatment of malaria.

9. **I. N. Dubin** (*introduced by* A. F. Coca), *Division of Pathology and Bacteriology, University of Tennessee College of Medicine*
   Studies in hypersensitivity in simian malaria.
10. **Paul de Gara and Henry P. Goldberg** (*by invitation*), *New York Hospital and Dept. of Pediatrics, Cornell University Medical College*
    Complement activity of sera from healthy and sick children.
11. **Christine E. Rice,** *Division of Animal Pathology, Science Service, Dominion Dept. of Agriculture, Animal Diseases Research Institute, Hull, Quebec, Canada*
    Paralleled use of the "direct" and "indirect" complement-fixation tests.
12. **Manfred M. Mayer and Charles C. Croft** (*by invitation*), *Dept. of Bacteriology, Johns Hopkins School of Hygiene and Public Health*
    On the kinetics of hemolysis by antibody and complement.

IMMUNOLOGY

Wednesday, March 17, 1:45 p.m.

ROOM 10, CONVENTION HALL

---

1. **Frank Maltaner and J. O. de Almeida** (*by invitation*), *New York State Dept. of Health, School of Medicine, University of Sao Paulo, Brazil*
   The effect of MG^{++} on the complementary and coagulative activities of blood serum.
2. **Daniel Boroff,** *Camp Detrick*
   A study on toxins and antigens of *S. dysenteriae.*
3. **Edgar E. Baker** (*by invitation*), **Ely Perlman and Walther F. Goebel,** *Rockefeller Institute for Medical Research and the Mt. Sinai Hospital, New York*
   The isolation and properties of specific antigens from variants of *Shigella Sonnei.*
4. **E. Singer, S. H. Wei and S. H. Hoa** (*introduced by* A. F. Coca), *Central Bacteriological Laboratory, National Institute of Health, Nanking, China*
   Immunological studies of cholera filtrates.
5. **Melvin Cohn** (*by invitation*) **and A. M. Pappenheimer, Jr.,** *Dept. of Bacteriology, New York University College of Medicine*
   Diphtheria toxin-antitoxin reaction in human antisera.
6. **James A. Harrison, Machteld E. Sano** (*by invitation*), **Elizabeth H. Fowler** (*by invitation*), **Robert H. Shellhamer** (*by invitation*), **and Carol A. Bocher** (*by invitation*), *Dept. of Biology, Temple University, and Temple University School of Medicine and Hospital*
   Toxicity for paramecia of sera from cancerous and non-cancerous persons.
7. **A. Packchanian,** *University of Texas School of Medicine*
   The production of anti-rabbit hemolysin (rabbit-erythrolysin) in sheep, and its value for complement fixation tests.
8. **J. McBroom Junge and S. M. Rosenthal** (*introduced by* A. F. Coca), *Division of Physiology, National Institute of Health, Bethesda, Md.*
   Effect of environmental temperature upon sulfadiazine therapy, body temperature and oxygen consumption in pneumococcus infection.
9. **Morris N. Green** (*introduced by* Stuart Mudd), *Dept. of Bacteriology, University of Pennsylvania School of Medicine*
   Studies on the mechanism of action of furacin (5-nitro-2-furaldehyde semicarbazone).
10. **M. G. Sevag and Joseph S. Gots** (*by invitation*), *Dept. of Bacteriology, University of Pennsylvania School of Medicine*
    Studies on pneumococcal enzymes involved in resistance to drugs.
11. **M. G. Sevag and Edward Steers** (*by invitation*), *Dept. of Bacteriology, University of Pennsylvania School of Medicine*
    The relation of tryptophane utilization to the mechanism of resistance to sulfonamides.

IMMUNOLOGY

Wednesday, March 17, 7:00 p.m.

ROOM 21, CONVENTION HALL

Symposia

I. SITE OF ANTIBODY FORMATION

1. **Philip D. McMaster,** LEADER (*by invitation*), *Rockefeller Institute for Medical Research, New York*
2. **Abraham White** (*by invitation*), *Yale University*
3. **Dan H. Campbell,** *California Institute of Technology, Pasadena*

II. ELECTRON MICROGRAPHY OF THE STRUCTURE OF BACTERIA AND VIRUSES

1. **Ralph W. G. Wyckoff,** LEADER, *National Institute of Health, Bethesda, Md.*
2. **Stuart Mudd,** *University of Pennsylvania School of Medicine*
3. **Geoffrey Rake,** *Squibb Institute for Medical Research, New Brunswick*
4. **Albert Claude** (*by invitation*), *Rockefeller Institute for Medical Research, New York*

Federation "Mixer"

Wednesday, March 17, 9:00 p.m.

ARENA, CONVENTION HALL

IMMUNOLOGY

Thursday, March 18, 9:00 a.m.

ROOM 10, CONVENTION HALL

---

1. **Elvin A. Kabat, Murray Glusman** (*by invitation*) **and Vesta Knaub** (*by invitation*), *Depts. of Neurology and Bacteriology, College of Physicians and Surgeons, Columbia University, and the Neurological Institute of New York*
   Immunochemical estimation of albumin and gamma globulin in normal and pathological cerebrospinal fluid.
2. **David Pressman and Geoffrey Keighley** (*introduced by* A. F. Coca), *Gates and Crellin Laboratories of Chemistry and the William G. Kerckhoff Laboratories of the Biological Sciences, California Institute of Technology, Pasadena*
   The zone of activity of antibodies as determined by the use of radioactive tracers.
3. **Herman N. Eisen and Albert S. Keston** (*introduced by* Robert C. Warner), *Depts. of Medicine and Chemistry, New York University College of Medicine*
   Measurement and study of immune precipitates by means of ultraviolet absorption spectroscopy. (Biochem.)
4. **Ellis Bolton, Charles Leone and Alan Boyden** (*introduced by* A. F. Coca), *Bureau of Biological Research and Dept. of Zoology, Rutgers University*
   A critical analysis of the performance of the photronreflectometer in the measurement of serological and other turbid systems.
5. **Henry P. Treffers and Katherine E. Yaw** (*by invitation*), *Section of Immunochemistry, Dept. of Bacteriology and Immunology, Yale University School of Medicine*
   A turbidimetric growth assay method for the determination of the relative bactericidal activities of sera.
6. **John P. Fox** (*introduced by* Peter K. Olitsky), *Laboratories of the International Health Division, Rockefeller Foundation, New York*
   The long persistence of *Rickettsiae orientalis* in the blood and tissues of infected animals.
7. **Edward C. Rosenow,** *Rare Metals Institute of the California Institute of Technology, Pasadena,* and *Longview Hospital, Cincinnati*
   Further studies on the production *in vitro* from bacteria of substances resembling "natural" agglutinins and precipitins.
8. **William Burrows,** *Dept. of Bacteriology and Parasitology, University of Chicago*
   Excretion of antibody in feces and urine and its absorption from the bowel.

IMMUNOLOGY

Papers Read by Title

1. **I. N. Dubin, J. D. Reese** (*by invitation*) **and Lois A. Seamans** (*by invitation*), *Division of Pathology and Bacteriology, University of Tennessee College of Medicine, and Dept. of Health and Safety, Tennessee Valley Authority*
   Attempt to produce protection against mosquitoes by active immunization.
2. **John P. Fox and Osler L. Peterson** (*introduced by* Peter K. Olitsky), *Laboratories of the International Health Division, Rockefeller Foundation, New York*
   Mode of action of thionine dyes in combatting experimental rickettsial infections of mice

DIRECTORY OF COUNCIL AND COMMITTEE MEETINGS AND SPECIAL FUNCTIONS NOT LISTED IN PROGRAM

Federation Executive Committee: Mandarin Room, Haddon Hall Hotel. Sunday March 14, 6:00–10:00 p.m.; Monday, March 15, 3:00–6:00 p.m.; Thursday, March 18, 9:00–11:00 p.m.

Federation Proceedings Control Committee: Room 1367, Haddon Hall Hotel. Monday, March 15, 9:00–11:00 a.m.

Council, Physiology: Room 1337, Haddon Hall Hotel. Sunday, March 14; Monday, March 15; Wednesday, March 17. *Times to be arranged.*

Council, Biochemistry: Room 1333, Haddon Hall Hotel. *Times to be arranged.*

Council, Pharmacology: Mandarin Room, Haddon Hall Hotel. Sunday, March 14, 10:00 a.m.; Monday, March 15, 10:00 a.m.; Wednesday, March 17, 10:00 a.m.

Council, Pathology: Room 1335, Haddon Hall Hotel. Sunday, March 14, 1:00 p.m.; Monday, March 15, 10:00 a.m.

Council, Nutrition: Green Room, Haddon Hall Hotel. Sunday, March 14, 1:00 p.m.; Monday, March 15, 9:00 a.m. and 1:00 p.m.

Council, Immunology: Room 1344, Haddon Hall Hotel. *Times to be arranged.*

Editorial Committee of the Annual Review of Physiology: Bakewell Room, Haddon Hall Hotel. Tuesday, March 16, 2:00 p.m.

Informal Discussion Group: "Nutritional Requirements of the Mouse." Room 15, Convention Hall. Wednesday, March 17, 7:30 p.m.

Society for Experimental Biology and Medicine Committee Meetings: Room 19, Convention Hall. Monday, March 15, 3:00 to 5:30 p.m.; Tuesday, March 16, 1:00 a.m. to 6:00 p.m.; Wednesday, March 17, 9:00 a.m. to 12:00 noon.

Poultry Nutrition Discussion Group: Room 15, Convention Hall. Monday, March 15, afternoon and evening.

Food and Nutrition Board, National Research Council. *a*) Committee on Nutrition Surveys: Room 19, Convention Hall. Monday, March 15, 9:00 a.m. *b*) Committee on Survey of Food and Nutrition Research. Room 19, Convention Hall. Monday, March 15, 10:30 a.m.

LUNCHEONS

City College of New York Alumni: Wednesday, March 17, 12:30 p.m. *Place to be arranged.*

Society for Experimental Biology and Medicine: Shelburne Hotel. *Time to be arranged.*

DINNER MEETINGS

1. **Circulation Group Dinner:** Wednesday, March 17, 6:30 p.m. (dinner 7:30 p.m.). *Place to be arranged.*
2. **Thudichum Society (Brain Chemistry Group):** Wednesday, March 17, 6:15 p.m. *Place to be arranged.*
3. **Metabolism Discussion Group:** Wednesday, March 17, 6:30 p.m. *Place to be arranged.*
4. **Editorial Board, Journal of Nutrition:** Thursday, March 18, 7:00 p.m. *Place to be arranged.*
5. **Editorial Board, Journal of Biological Chemistry:** Thursday, March 18, 7:00 p.m. Haddon Hall Hotel.
6. **Editorial Board, Biochemical Preparations:** Thursday, March 18, 6:30 p.m. Haddon Hall Hotel.
7. **Society for Experimental Biology and Medicine:** Shelburne Hotel. *Time to be arranged.*

INDEX OF AUTHORS

A

Abell, L. L. 371
Abendroth, M. 370
Abrams, A. 342
Abreu, B. E. 364
Achenbach, P. 353
Acheson, G. H. 328
Acosta, R. R. 352
Adams, B. J. 358, 366
Adams, M. 363
Adler, M. 351
Adolph, E. F. 319, 338
Agner, K. 368
Ahlquist, R. P. 353
Albanese, A. A. 349, 376
Albaum, H. G. 343
Algire, G. H. 314
Allen, F. M. 371
Allen, J. G. 324
Allen, S. C. 364
Allen, T. H. 337
Allgeier, A. M. 374
Allison, J. B. 349
Alonso, L. 363
Alper, C. 344
Altman, K. I. 336, 346
Alture-Werber, E. 356
Amberson, W. R. 334
Ambrose, A. M. 359, 364
Ames, A., III 319
Ames, S. R. 342
Ampil, D. 318, 319
Ananenko, E. 355
Anderson, A. 322
Anderson, A. A. 376
Anderson, C. E. 330, 332
Anderson, E. P. 373
Anderson, H. H. 364
Anderson, J. A. 349
Andrews, J. C. 352
Anderson, J. M. 336
Anderson, M. M. 333, 340
Anderson, R. S. 334
Anderson, W. L. 322
Anker, H. S. 341
Anslow, W. P., Jr. 331, 332
Archibald, R. M. 352, 357
Armstrong, W. D. 341
Artom, C. 343
Asenjo, C. F. 347
Ashby, W. 349
Attyah, A. M. 329
Austin, G. M. 333
Axelrod, J. 360
Ayer, J. L. 332

B

Bach, L. M. N. 331
Bachelor, A. L. 365
Bader, M. E. 319
Baez, S. 315, 340
Bailey, C. A. 377
Baily, J. 359
Bain, J. A. 349
Baker, E. E. 378
Baker, R. D. 371
Bakos, A. C. P. 336
Baldwin, E. deF. 330
Balistocky, M. 324
Bardwell, K. 327
Barger, A. C. 320
Barker, S. B. 325
Barnes, R. B. 346
Barnes, T. C. 326, 363, 367
Barnes, T. R. W. 365
Barnett, H. L. 315, 332, 336
Barrett, M. J. 328
Barrett, W. 330
Barron, D. H. 315, 318
Bartlett, G. R. 338
Bartlett, P. 349
Bass, A. D. 361
Batten, W. 315
Batterman, R. C. 356
Baumann, C. A. 345, 372
Baumberger, J. P. 327
Baxter, J. H. 367
Baylor, M. B. 335
Bazett, H. C. 317, 319
Beach, F. F. 375
Beach, J. 325
Bean, J. W. 326
Bean, R. S. 350
Beard, H. H. 352
Beatty, C. H. 324
Beck, O. 373
Beckman, H. 365
Beecher, H. K. 354, 356
Begany, A. J. 360
Behrmann, V. G. 332
Belding, H. S. 319
Bell, F. K. 363
Bell, M. 356
Beman, F. M. 327
Bender, M. B. 333
Benditt, E. P. 370
Benton, J. G. 363, 364
Berg, C. P. 344
Berkson, J. 351
Bernheim, F. 356, 373
Berry, I. M. 337
Berry, R. G. 335
Beshore, D. L. G. 320
Besman, L. 373
Beutner, R. 326
Bevans, M. 371
Beyer, K. H. 356
Bhattacharya, B. K. 365
Bieter, R. N. 359
Bing, R. J. 329
Binkley, F. 348
Bird, H. R. 373
Bird, J. G. 360
Bird, O. D. 347
Bird, R. M. 349
Birmingham, M. K. 349
Birnie, J. H. 338
Bixby, E. W. 317, 323
Black, S. 341
Black, S. P. W. 331
Bliss, A. F. 327
Bloch, K. 341
Block, S. 338
Bobb, J. R. R. 336, 359
Bocher, C. A. 378
Bodansky, O. 343, 363
Bodine, J. H. 327
Boehne, J. W., III 374
Bollman, J. L. 323, 343, 367, 371
Bolomey, A. A. 315, 332
Bolton, E. 379
Bondy, P. K. 330
Bonnycastle, D. D. 359
Booker, W. M. 315, 358
Boone, B. R. 329
Borchardt, P. R. 316
Borek, E. 347
Borison, H. L. 317
Boroff, D. 378
Boroughs, E. D. 374
Borsook, H. 346, 348
Bowen, W. J. 334, 338
Bowers, R. V. 334
Bowman, D. E. 350
Boyarski, L. L. 322
Boyd, E. M. 356
Boyden, A. 379
Boyle, D. 333
Boyles, P. W. 369
Bozler, E. 329
Bradbury, J. T. 321
Bradley, S. E. 330
Brand, F. C. 343
Bratton, A. C., Jr. 358
Brauer, R. W. 364
Braun, H. A. 355
Breed, E. F. 315
Brendler, S. 334
Brewer, J. H. 376
Brewer, N. R. 330
Bridge, E. M. 346
Briggs, A. P. 329
Briggs, G. M. 373
Brink, F. 319, 322, 327
Briody, A. B. 377
Britton, S. W. 327
Brobeck, J. R. 320, 328
Brodie, B. B. 354, 360
Brody, D. A. 329, 338
Broh-Kahn, R. H. 325, 330, 338
Bronk, D. W. 319, 322
Brookhart, J. M. 331
Brooks, C. McC. 316
Brooks, V. B. 322
Brow, L. 373
Browe, J. H. 375
Brown, E. B. 320
Brown, F. 338
Brown, G. E., Jr. 321
Brown, R. A. 347
Brown, R. V. 360
Brown, W. 338
Brown, W. E. 321
Brozek, J. 317
Brucer, M. 320
Bruger, M. 352
Brumbaugh, M. A. 358
Bruner, H. 355
Bruns, P. 318
Brush, M. 374
Bucher, G. R. 322
Bucker, M. L. 374
Buckingham, M. 377
Burbridge, T. N. 365
Burge, W. E. 326
Burk, D. 342, 351
Burke, J. C. 357
Burks, A. L. 364
Burnett, C. H. 354
Burnett, R. C. 366
Burnett, W. T. 372
Burney, T. E. 377
Burr, H. S. 338
Burr, M. M. 351
Burris, R. H. 344
Burrows, W. 379
Buschke, W. 327
Byers, S. O. 338

C

Cain, O. 323, 332
Caldwell, A. 352
Calkins, E. 347
Callender, S. 370
Cameron, A. 353, 360, 363, 365
Camp, W. J. R. 358
Campbell, D. H. 377, 378
Campbell, G. S. 320
Canepa, J. F. 330
Cannan, R. K. 341
Cannon, P. R 314
Cantarow, A. 320, 331, 333
Canzanelli, A. 325, 330
Capo, L. R. 359
Capps, R. T. 361
Carlisle, E. M. 375
Carlo, J. 333
Carlson, F. D. 322
Carlson, L. D. 315, 338
Carlson, R. M. 360
Carmichael, E. B. 367
Carpenter, C. P. 332
Carr, C. J. 333, 354
Carroll, R. T. 368
Carter, C. E. 350
Casarett, G. W. 336
Casas, C. B. 333
Cassels, W. 360
Cassidy, G. H. 346
Caster, W. O. 375
Castillo, J. C. 362
Catchpole, H. R. 348
Cavett, J. W. 351
Ceithmal, J. 350
Cerecedo, L. R. 374, 376
Chadbourn, W. A. 359
Chadwick, L. E. 327, 328
Chandler, B. F. 334
Chandler, J. P. 348
Chang, T. S. 338
Chanutin, A. 342
Chapin, J. L. 338
Chapman, C. B. 317
Chapman, E. 376
Chargaff, E. 343
Charipper, H. A. 325
Charrington, M. E. 369
Chase, H. F. 365
Chatfield, P. O. 317
Chavarria, A. P. 365

Chen, G. 358
Chen, J. Y. P. 359
Chen, S. D. 373
Cheng, A. L. 375
Chenoweth, M. B. 353, 360, 366
Chernoff, H. M. 316
Child, G. P. 338, 364
Chin, Y. 365
Chinard, F. P. 325
Chinn, B. 334
Chow, B. F. 344, 349
Christensen, H. E. 354
Christensen, H. N. 344
Chu, N. 363
Ciethaml, J. 350
Cisler, L. E. 340
Cizek, L. J. 331
Clagett, O. T. 332
Clark, G. 317, 318
Clark, W. G. 318
Clark, W. M. 348
Clarke, R. W. 315
Claude, A. 378
Clayton, R. 375
Close, J. R. 334
Cohen, P. P. 342, 346
Cohn, M. 378
Cohn, T. 368
Cole, V. V. 354, 358, 359
Collings, W. D. 323
Collins, D. A. 330
Collins, G. L., Jr. 346
Compton, D. W. 354
Comroe, J., Jr. 355
Connelly, C. M. 322
Conway, A. C. 354
Cook, D. L. 362
Cooke, R. C. 328, 375
Coon, J. M. 354, 357, 361, 366
Copeland, J. E. 356
Copley, A. L. 369
Copp, D. H. 337
Coppedge, R. L. 369
Corcoran, A. C. 326, 332
Corey, E. L. 327
Cornatzer, W. E. 343
Corson, S. A. 323, 332
Cort, J. H. 315
Coryell, M. N. 375
Cotlove, E. W. 363, 364
Co Tui, F. 364, 365
Cotzias, G. C. 325
Cournand, A. 329, 336
Cowdery, P. C. 364
Cowgill, G. R. 372
Cox, A. A. 318
Craig, F. N. 318
Craig, L. C. 346
Crandall, W. A. 351
Cranston, E. M. 359
Craver, B. 330, 353, 360, 363
Cregar, M. 327
Crismon, J. M. 338
Critchfield, F. H. 332, 348, 373
Croft, C. C. 378
Crookshank, H. R. 344
Crosley, A. P., Jr. 315
Cross, N. 348
Csonka, F. A. 342
Cubiles, R. 343
Cucci, M. R. 362
Cuckler, A. C. 359
Curnen, E. C. 377
Curtis, J. M. 345
Cusick, G. 332
Custer, J. H. 342

D

Dackerman, M. E. 350
DaCosta, E. 375
Damron, C. M. 373
D'Angelo, S. A. 325
Danowski, T. S. 324, 337
Daum, K. 317
Daus, M. A. 345
David, N. A. 355
Davidow, B. 361, 362
Davies, P. W. 322
Davis, B. D. 377
Davis, F. 373
Davis, G. D. 327
Davis, H. G., Jr. 371
Davis, J. E. 365
Davis, M. E. 365
Davis, P. L. 360
Davis, R. E. 330
Davis, S. K. 334
Davis, V. I. 349
Davis, W. L. 377
Davison, C. 355
Day, B. 376
Day, H. G. 372
Day, M. A. C. 325
Day, P. L. 347
Dayrit, C. 363, 364
de Almeida, J. O. 378
Deasy, C. L. 346
DeBiase, S. 344, 349
de Bodo, R. C. 324, 325
de Duve, C. 346
Deering, R. 335
de Gara, P. F. 378
de Grandpre, R. 325
deGuisti, D. 359
De Meio, R. H. 320
Denslow, J. S. 334
Denton, J. E. 356
Denton, R. W. 330
DeRitter, E. 376
De Robertis, E. 319
Dethier, V. G. 327, 328
Devel, H. J., Jr. 375
Deyrup, I. J. 336
Dierks, F. H. 377
Dill, W. A. 363
DiPalma, J. R. 328, 329
Dische, Z. 318, 343
Dixon, F. 371
Dobbiner, K. 345
Dobson, H. L. 316
Doisy, E. A. 345, 351
Dontigny, P. 325
Dougherty, I. 325
Doull, J. 361
Dow, P. 328
Downs, W. L. 362
Drabkin, D. L. 348, 351
Drake, C. G. 338
Drake, C. V. 331
Draper, W. B. 317, 320, 335
Dreisbach, R. H. 363
Dreyer, N. B. 367
Drill, V. A. 361, 365
Drinker, C. K. 336
Driver, R. L. 329
Droogleever-Fortuyn, J. 335
Dryer, R. L. 323, 352
Dubach, R. 370
Dubin, A. 367
Dubin, I. N. 378, 379
Dubnik, C. S. 375
Dubnoff, J. W. 346, 348
DuBois, K. P. 361, 365
Duca, C. J. 357
Dulaney, A. D. 377
Dumm, M. E. 372
Dunn, T. B. 375
Dusse, E. V. 335
Dutcher, J. D. 351
Dutcher, R. A. 372
du Vigneaud, V. 348
Dyniewicz, H. M. 367
Dziemian, A. J. 321

E

Eadie, G. S. 356
Eagle, H. 355
Eaton, M. D. 377
Eccles, J. C. 316
Eckenhoff, J. E. 329, 336, 363
Ecker, E. E. 377
Eder, H. A. 325
Edwards, C. T. 322
Edwards, L. E. 322
Egner, W. 324
Ehrich, W. E. 368
Eiseman, A. J. 356
Eisen, H. N. 371, 379
Eisenberg, L. 319
Elam, J. O. 320, 329
Eldredge, D. H., Jr. 335
Elion, G. B. 347
Elkins, E. 350
Elkins, E. C. 332
Ellinger, G. F. 329
Elliott, H. W. 365
Elliott, K. A. C. 349
Elliott, K. J. 373
Elliott, W. H. 351
Ellis, D. 320
Ellis, S. 356
Elvehjem, C. A. 314, 373
Elwell, L. H. 326
Emerson, G. A. 347, 353, 354
Engel, L. L. 345
Engel, R. W. 372
Engelmann, T. 319
Ensor, C. 358
Eppright, M. 375
Ercoli, N. 356
Erdös, T. 334
Escher, D. J. W. 326, 328, 333
Essenberg, J. M. 331
Evans, A. S. 377
Evans, E. A., Jr. 342
Evans, J. D. 349
Evans, L. 330
Everett, F. G. 365
Everett, G. M. 357
Eversole, W. J. 338
Everson, G. 375
Ewing, P. L. 353, 354

F

Fairfield, J. 338
Faraco, E. 364
Farah, A. 360
Faribault, C. 333
Farrar, G. E., Jr. 364
Fassett, D. W. 360
Faust, C. E. 343
Fearing, D. F. 336
Fearney, F. 343
Feigen, G. A. 315
Feinstein, B. 334
Feld, E. A. 321
Feldman, B. 344
Fellers, F. X. 336
Felton, L. D. 376
Fenton, P. F. 372
Ferguson, J. H. 369, 370
Ferguson, J. K. W. 361
Ferry, J. D. 352
Fetcher, E. S. 319
Fiala, S. 342
Field, J. 322
Finch, C. A. 374
Finch, E. P. 370
Finkelstein, M. 363
Finnegan, J. K. 354
Fischer, C. J. 321
Fischer, Ernst 334
Fischer, G. 336
Fishback, H. R. 340
Fishberg, E. H. 344
Fishman, W. H. 346
Fisken, R. A. 370
Fitzgerald, D. B. 356
Fitzgerald, L. R. 327
Fitzhugh, O. G. 355
Fitzpatrick, T. B. 347
Flinker, M-L. 336
Flock, E. V. 343, 367, 371
Fluharty, R. G. 358
Foa, P. P. 337
Folch, J. 342
Follis, R. H., Jr. 368
Fong, C. T. O. 346
Foltz, E. L. 329, 363
Forbes, J. C. 343
Forman, C. 368
Forsham, P. H. 333
Forster, F. M. 335
Foster, E. 332
Foster, R. H. K. 368
Foster, W. C. 351
Fowell, D. M. 329
Fowler, E. H. 373
Fowler, W. S. 317
Fox, J. P. 379
Fradkin, R. 359
Fraenkel-Conrat, H. 350
Frame, B. 361
Frank, K. 331
Franklin, A. L. 347, 353, 374
Frantz, J. A. 323
Fraser, D. 335
Fraser, W. D. 346
Frazier, E. I. 372
Freedman, A. M. 320, 338, 357
Freeman, L. W. 331
Freeman, S. 328, 338
Freese, H. B. 362
French, D. M. 315, 358
Frenchman, R. 374
Frey, J. S. 315
Freyburger, W. 359
Freyhan, F. A. 323
Frias, J. E. G. 359

Frick, J. D. 355
Friedberg, F. 346
Frieden, E. 351
Friedgood, C. E. 344
Friedkin, M. E. 350
Friedman, M. 338
Friedman, M. H. F. 338, 339, 368, 370
Friedman, S. 338
Fruton, J. S. 350
Fugo, N. W. 365
Fuhrman, F. A. 338
Fukushima, D. K. 345
Fuller, F. D. 339
Fulton, J. F. 335
Furchgott, R. F. 329, 340
Furman, R. H. 369
Furth, J. 370
Fusco, M. 327
Futcher, P. H. 315

G

Gaddy, C. G. 332
Gaebler, O. H. 349
Gaines, D. S. 347
Galdston, M. 317
Gale, R. G. 330, 369
Gallant, D. L. 351
Gamble, J. L., Jr. 327, 339
Gant, V. A. 358
Gantt, W. H. 317, 331
Garb, S. 353, 360
Gardner, R. A. 354
Garzoli, R. F. 371
Gast, J. H. 316
Gattone, V. 338
Gauer, O. 327
Gaunt, R. 338
Gawron, O. 356
Geiger, A. 321
Geiman, Q. M. 351
Geissman, T. A. 338
Gelfan, S. 327
Gemmill, C. L. 348, 365
Genther, C. S. 359
Geraci, J. E. 314, 317, 320
Gerheim, E. B. 369, 370
German, H. L. 376
Germuth, F. G., Jr. 355
Gersh, I. 366, 369
Gesell, R. 315
Geyer, R. P. 373
Giarman, N. J. 360
Gilder, H. 348
Gimble, A. I. 355
Gjessing, E. C. 342
Glaser, K. 357
Glasser, O. 332, 340
Glazko, A. J. 363
Glover, M. 360
Glusman, M. 379
Goddard, L. 363
Godfrey, E. 361
Goebel, W. F. 378
Goettsch, M. 376
Goetzl, F. R. 334
Gold, H. 363, 364
Gold, J. J. 329
Goldat, S. 328
Goldbaum, L. R. 358
Goldberg, E. 359
Goldberg, H. 324
Goldberg, H. P. 378
Golden, A. 370, 371
Goldensohn, E. S. 335
Goldman, E. 352
Goldman, M. L. 315, 321, 326
Goldsmith, G. A. 372, 375
Goldstein, A. 361
Goldstein, M. J. 316
Goldthwait, D. A. 319
Gollan, F. 320
Gollub, M. C. 350
Golub, O. J. 377
Gomez, D. M. 315
Gongora, J. 318
Goodman, L. S. 314
Goodwin, M. P. 345
Gordon, A. S. 325
Gordon, E. B. 354
Gordon, F. B. 370
Gordon, G. S. 364
Gosselin, R. E. 339
Goth, A. 356
Gots, J. S. 378
Gould, R. G. 341
Graham-Service, D. M. 334
Grant, F. C. 333
Grant, R. 323
Grant, W. C. 332
Graybiel, A. 334
Greco, A. 352
Green, A. A. 320
Green, D. M. 332, 360
Green, H. D. 315, 323, 332, 359
Green, M. N. 378
Greenberg, D. M. 346, 352
Greenberg, L. D. 344, 369
Greenberg, S. M. 363
Greenman, L. 324, 337
Greenstein, J. P. 347
Greep, R. O. 321, 324
Greif, R. L. 325
Greisheimer, E. M. 320
Grenell, R. G. 322, 338
Griffin, A. C. 342
Griffin, G. E. 345
Griffith, L. E. 345
Griffith, R. S. 319
Griffith, W. H. 349
Grindlay, J. H. 367
Grisolia, S. 346
Groat, R. A. 339
Groff, A. E 330
Groom, D. L. 320
Groschke, A. C. 373
Gross, E. G. 355, 365
Grossman, M. I. 322, 328, 330
Grubbs, R. C. 347
Gruber, C. M. 357
Gruhzit, O. M. 370
Grundfest, H. 318, 319, 326
Guest, M. M. 327
Guild, L. 328
Guild, R. 325, 330
Gunning, R. L. 330
Guo, K. 377
Guthrie, C. C. 339
Gutmann, H. R. 350
Guttman, R. 319

H

Haag, H. B. 354
Haag, J. R 375
Haagen-Smit, A. J. 346
Haber, C. 322
Hack, M. H. 337
Hafkenschiel, J. H. 329, 336
Hafkesbring, R. 316
Hagan, E. C. 361, 362
Hahn, P. F. 348, 371
Haig, C. 333
Haig, E. M. 333
Haines, W. J. 345
Haldi, J. 317
Hale, E. 322
Hale, W. M. 376
Hall, J. F. 319
Hall, V. E. 323
Hall, W. K. 344
Hallenbeck, G. A. 332
Hambly, A. S., Jr. 365
Hambourger, W. E. 362
Hamilton, W. F. 329
Hammett, F. S. 339
Hampson, J. L. 335
Handelsman, J. C. 329
Handler, P. 344, 372, 373
Handley, C. A. 365
Hanke, M. E. 342
Hanlon, L. W. 363, 364
Hansen, A. E. 354, 373
Hansen, E. L. 365
Hansen, N. 365
Hanson, H. T. 350
Hanson, M. E. 328
Haour, P. 333
Hara, M. 336
Hardenbergh, E. 336
Hardman, B. S. 338
Hardy, J. D. 328, 331, 335
Hare, K. 315, 332, 336
Hare, R. S. 332
Harkness, D. M. 344
Harmel, M. H. 322
Harris, J. S. 344
Harris, R. S. 328, 374, 375
Hart, W. M. 334
Harte, R. A. 349
Hartline, H. K. 319, 333
Harrison, J. A. 378
Harvey, A. McG. 328
Harvey, R. B. 315
Haskell, B. F. 368
Hassett, C. C. 327
Hastings, A. B. 341
Haukohl, R. S. 367
Hauser, P. J. 324
Hawk, W. D. 368
Hawkins, J. E., Jr. 356
Hawkins, M. 353
Hays, H. 361, 363
Hazleton, L. W. 361
Hearon, J. 342
Hechter, O. 333
Hege, J. R., Jr. 315
Hegsted, D. M. 372, 374
Heidelberger, M. 377
Heilbrunn, L. V. 324
Heinle, R. W. 347
Heller, C. G 325
Hellman, L. 326, 333
Hellman, L. M. 318
Helmer, O. M. 326
Hemingway, A. 320
Hendley, C. D. 335
Hendricks, E. L. 354
Henneman, E. 334, 335
Henry, J. P. 327, 337, 339
Henschel, A. 317
Heppel, L. A. 364
Herrick, J. F. 323
Herriott, R. M. 346
Herrmann, R. G. 365
Herrold, E. 353, 361, 365
Hertzman, A. 323, 332
Hess, W. C. 344
Hesselbach, M. 342
Hiatt, E. P. 321
Hier, S. W. 344
Hiestand, W. A. 339
Highman, B. 367
Hill, E. 343
Hill, R. M. 375
Hillier, J. 346
Himmelstein, A. 329, 336
Himwich, H. E. 320, 338, 357
Himwich, W. A. 348
Hine, C. H. 354, 365
Hirshberg, E. 376
Hirschmann, F. B. 345
Hirschmann, H. 345
Hirshfeld, A. 343
Hitchcock, F. A. 320, 327, 347
Hitchings, G. H. 347
Hoa, S. H. 378
Hoagland, H. 333
Hodes, R. 317, 319, 339
Hodge, H. C. 362
Hoffman, W. S. 374
Hoffmaster, T. 366
Hogan, A. G. 374
Holaday, H. A. 357
Holland, H. L. 365
Holland, O. L. 355
Hollander, F. 322
Hollander, V. 365
Holman, R. L. 371
Holman, R. T. 343
Holmes, J. H. 331
Holmes, J. O. 373
Holmquist, G. 361, 363
Holt, L. E., Jr. 349, 376
Holtkamp, D. E. 375
Homburger, F. 342
Hopkins, A. L. 327
Hoppe, J. O. 356
Hopper, S. H. 358, 359
Horecker, B. L. 350
Horlick, L. 326
Horn, M. J. 352
Horvath, P. N. 323
Horvath, S. M. 323, 339
Horwitz, O. 329
Horwitz, S. A. 317
Houchins, R. 347
Houck, C. R. 339
Houghton, B. C. 347
House, F. 376
Hove, E. L. 373
Howe, E. E. 345
Howie, D. L. 359
Hsien-gieh, S. 343
Huf, E. G. 334
Hulpieu, H. R. 354, 358, 359
Humm, F. D. 318
Humphrey, M. J. 372
Humphreys, E. M. 366
Hunt, C. C. 353, 366
Hunter, F. E., Jr. 350

Hunter, J. A. 320
Hurn, M. 369
Hutchens, J. O. 327
Hutcheon, D. E. 354
Hutt, B. K. 339
Hwang, K. 329, 365

I

Ingle, D. J. 333
Ingraham, R. C. 324
Inman, V. T. 334
Irvin, E. M. 342
Irvin, J. L. 342
Isbell, H. 356
Ivy, A. C. 328, 329, 330, 337, 355, 360
Iwamoto, H. K. 363

J

Jackson, E. B. 370
Jacobs, H. 327, 337
Jaeger, M. 363
Jahan, I. 332
Jahn, T. L. 340
Jailer, J. W. 345
James, H. P. 365
Jamison, R. 360
Janton, O. H. 339
Jasper, H. H. 335
Javert, C. T. 331
Jellinek, E. M. 358
Jenney, E. H. 355, 366
Jensen, D. 369
Jochim, K. E. 332
Joffe, M. 327
Johnson, A. D. 332
Johnson, F. H. 335
Johnson, R. E. 352
Johnson, R. M. 372
Johnson, W. H. 367
Johnston, C. D. 362
Johnston, F. A. 374
Johnston, M. W. 325
Johnstone, H. G. 364
Jones, D. B. 352
Jones, H. B. 333
Jones, H. P. 360
Jones, H. W., Jr. 357
Joseph, N. R. 323, 339
Jukes, T. H. 347, 353, 374
Jung, F. T. 340
Jungck, E. S. 325
Junge, J. M. 378

K

Kabat, E. A. 379
Kade, C. F., Jr. 349
Kaiser, R. 356
Kalnitsky, G. 350
Kamin, H. 344
Kaplan, A. 325
Kaplan, E. 339
Kaplan, N. O. 341
Karczmar, A. G. 354
Karel, L. 357, 362
Karp, A. 351
Karpovich, P. V. 317
Karr, N. W. 354, 366
Karstens, A. 327, 337
Kartin, B. 316
Katsh, G. F. 325
Katz, J. 323, 352
Katz, L. N. 326, 333
Kaufman, W. 316
Kaulbersz, J. 322
Kaunitz, H. 369
Keating, F. R., Jr. 351
Keighley, G. 346, 379
Kelley, B. 372
Kelly, A. R. 358, 366
Kelsey, F. E. 377
Kelsey, M. P. 316
Kemph, J. A. 327
Kendall, F. E. 371
Kennedy, E. P. 341
Kensler, C. J. 358
Keston, A. S. 341, 342, 365, 371, 379
Ketron, K. D. 376
Kety, S. S. 321
Keys, A. 317, 375
Keyser, G. F. 357
Kiang, S. P. 325
Kielley, W. W. 343
Kik, M. C. 376
Killough, J. H. 371
Kimura, K. K. 357
King, E. N. 339
King, J. T. 333
King, T. O. 353
Kinney, T. D. 374
Kirchhof, A. C. 366
Kirk, J. S. 324
Kisch, B. 316, 339
Klein, J. R. 349
Klein, M. 376, 377
Kleitman, N. 319
Klug, H. L. 352
Knaub, V. 379
Knox, N. 364
Kochakian, C. D. 318, 320
Koehler, A. E. 343
Koenig, H. 336
Koenig, R. 336
Kohlstaedt, K. G. 326
Komarov, S. A. 322
Koppanyi, T. 354
Kornberg, A. 343
Korr, I. M. 316
Kottke, F. J. 315
Kozloff, L. M. 342
Kozoll, D. D. 374
Kramer, B. 352, 373
Kramer, M. L. 363
Kramer, W. 341
Krampitz, L. O. 341
Krantz, J. C., Jr. 354
Krause, R. F. 375
Kraushaar, O. F. 321
Krayer, O. 360
Krebs, E. G. 346
Krehl, W. A. 372, 374
Kriss, J. P. 315
Krueger, A. P. 368
Kubicek, W. G. 315
Kuether, C. A. 376
Kuffler, S. W. 328
Kuiken, K. A. 351, 374
Kun, E. 368
Kuna, S. 356
Kunkel, H. O. 375
Kuntz, R. E. 371
Kunze, D. C. 336, 359
Kunze, F. M. 355
Kupperman, H. S. 355
Kwit, N. T. 363, 364

L

LaBelle, A. 318, 363
LaBelle, C. W. 362
Lachman, P. O. 330
LaForge, M. 365
Lambert, E. H. 316
Lameck, W. I. 375
Lampson, G. P. 352
Landis, E. M. 320
Lane, A. 328
Larrabee, M. G. 317, 319
Larson, P. S. 354
Laskey, M. 360
Laskin, S. 362
Lauber, F. U. 322
Laug, E. P. 355, 362
Lauson, H. D. 325
Lawson, H. D. 315
Lawson, J. D. 338
Leake, C. D. 366
Leary, J. S. 360
Leathem, J. H. 318
Leduc, E. H. 373
Leduc, J. 325
Lee, A. S. J. 352
Lee, J. M. 348
Lee, L. E. 358
Lee, M. E. 339
Legallais, F. Y. 314
Lehman, R. A. 355
Lehninger, A. L. 341, 350
Lehr, D. 359, 366
Leimdorfer, A. 316, 339, 367
Lein, M. 349
Leiner, G. 328
Leiter, L. 326, 328, 333
Leland, M. L. 356
LeMessurier, D. H. 335, 339
Lemley, J. M. 369
Lenel, R. 332
Leonards, J. R. 351
Leong, G. F. 327
Leone, C. 379
Lerner, A. B. 347
Leslie, S. 330
Lettvin, J. Y. 316
Levedahl, B. H. 345
LeVeen, H. H. 374
Levin, L. 345
Levine, M. D. 371
Levine, R. 332, 337
Levine, V. E. 352
Levy, M. 341, 342
Lewey, F. H. 331, 333
Lewis, J. C. 366
Lewis, J. H. 369
Lewis, L. A. 326
Lewis, M. N. 356
L'Heureux, V. 352
Li, M. C. 339
Libet, B. 322
Liddle, G. W. 364
Lieberman, S. 345
Lief, P. A. 354
Ligon, E. W. Jr. 360
Lindberg, O. 343
Lindenbaum, A. 341
Linegar, C. R. 357, 360
Lineweaver, H. 350
Ling, G. 334
Lipmann, F. 341
Lipner, H. J. 325
Lippman, R. W. 339
Lipsett, M. 342
Lipton, E. 357
Lisi, A. G. 354
Litchfield, J. T., Jr. 358, 363
Little, J. M. 315, 320, 332
Littman, A. 322
Lium, R. 367
Livingston, R. B. 335
Lloyd, D. P. C. 316
Lobb, A. 332
Loeb, P. 357
Loew, E. R. 353
Loewe, L. 356
Loewe, S. 367
London, I. M. 344
Loomis, T. A. 356
Loosli, C. G. 370
Lotspeich, W. D. 332
Love, J. 367
Love, L. 319
Loveless, M. H. 377
Lovseth, L. 360
Lowell, F. C. 377
Lowell, V. F. 358
Lowry, P. H. 346
Lu, G. 366
Luck, J. M. 342
Ludewig, S. 342
Ludwig, A. S. 337
Luduena, F. P. 356
Lugovoy, J. K. 350
Lukens, F. D. W. 321
Lunbaek, K. 324
Lusky, L. M. 356
Lyman, C. M. 351, 374

M

Macy, I. G. 375
Macht, D. I. 318, 339, 354, 366
Macht, M. B. 319, 339
MacKay, D. 376
Mackenzie, A. 363, 365
Mackenzie, C. G. 348
MacNichol, E. F. 333
MacVicar, R. 344
Madden, R. A. 360
Maddock, C. L. 360
Maddock, S. 367
Maddock, W. O. 325
Magee, M. 339
Magnes, J. 321
Magoun, H. W. 331
Maher, P. J. 327
Maison, G. L. 356
Maitland, B. 346
Malcolm, J. L. 316
Maloney, J. 355
Maltaner, F. 379
Malton, S. D. 354
Maluf, N. S. R. 339
Manery, J. F. 340
Mangun, G. H. 353
Mann, F. D. 360

Mann, G. V. 373
Maren, T. H. 359
Markee, J. E. 316
Marrazzi, A. S. 353
Marsh, D. F. 357
Marshall, J. 325, 339
Marsters, R. W. 351
Martin, A. W. 315, 334, 338
Martin, C. J. 323
Martin, E. M. 339
Martin, M. J. 339
Marx, W. 342
Marzulli, F. N. 318
Mason, H. L. 345
Masters, R. E. 336
Mathieson, D. R. 369
Maver, M. E. 352
Mayer, J. 372
Mayer, M. M. 377, 378
Maynard E. A. 362
Mayock, R. L. 329
Mazur, A. 315
McBride, T. 368
McCall, W., Jr. 336
McCawley, E. L. 357
McClendon, J. F. 351, 376
McClosky, W. T. 359
McConahey, W. C., Jr. 351
McCulloch, W. S. 335
McFee, R. 336
McGinty, D. A. 363
McIntyre, A. K. 316
McKee, A. P. 376
McKee, M. 347
McKee, R. W. 351
McKey, B. V. 375
McKibbin, J. M. 349
McLain, P. L. 337
McMahon, T. M. 327
McManus, J. F. A. 369
McMaster, P. D. 378
McMeekin, T. L. 342
McNamara, H. 315, 332, 336
McNeil, G. 374
McOmie, W. A. 366
Mead, S. 317
Mecham, D. K. 352
Mederos, L. O. 316
Meeham, H. 337
Meier, R. 343
Meister, A. 347
Mellette, H. 327
Melnick, J. L. 376
Melton, F. M. 340
Melville, K. I. 360
Member, S. 352
Mendelow, H. 343
Mendez, R. 361
Meneely, G. R. 369
Meng, H. C. 328
Menkin, V. 370
Menzel, A. E. O. 351
Merlis, S. 352
Merriam, H. 374
Merrow, S. 375
Mettler, F. A. 335
Meyer, B. J. 357
Meyer, K. 346
Meyer, K. A. 358
Meyer, W. 371
Meyers, J. D. 324
Meyerhof, O. 343
Michaelis, L. 348
Michie, A. 315
Michie, C. R. 329
Michie, K. 315
Michiels, P. M. 362
Mickelsen, O. 375
Milhorat, A. T. 339, 373
Miller, A. T., Jr. 317
Miller, C. P. 368
Miller, F. A. 345
Miller, L. C. 355, 356
Miller, L. L. 331, 344
Miller, O. B. 318
Miller, O. N. 350
Miller, R. C. 372
Miller, R. N. 339
Millman, N. 317
Mills, M. B. 373
Minatoya, H. 325
Minckler, J. 317
Minnich, V. 370
Mirone, L. 374, 376
Mirsky, I. A. 325, 325, 330, 338
Modell, W. 363, 364
Moe, G. K. 354, 359
Moersch, G. W. 347
Molano, F. A. 315, 358
Molitor, H. 356
Molland, J. 359
Monaghan, E. A. 358
Monica, E. S. D. 342
Montgomery, G. E., Jr. 314, 317, 320
Moore, B. 338
Moore, C. V. 370
Moore, J. C. 337, 338
Moore, M. C. 335
Moore, M. E. 376
Moore, S. 341
Moreau, L. 324
Morehouse, M. G. 375
Morris, H. P. 375
Morris, R. E. 331
Morrison, J. L. 358
Mortarotti, T. G. 364
Morton, G. M. 330
Moses, F. E. 368
Moss, W. G. 325
Motley, H. L. 336
Mountcastle, V. 334
Moxon, A. L. 352
Mudd, S. 378
Mulholland, J. H. 364, 365
Müller, O. H. 321
Munro, F. L. 369
Munro, M. P. 369
Munsell, H. E. 328, 374
Muntwyler, E. 344, 345
Mushett, C. W. 322, 347
Musser, R. 354
Myers, V. C. 351

N

Nachmansohn, D. 321, 326, 341
Nahum, L. H. 316
Najjar, V. A. 346
Nakamura, K. 348
Natelson, S. 350, 352
Nawrocki, M. F. 349
Neilands, J. B. 372
Nelson, A. A. 355, 361, 367
Nelson, D. 339
Nelson, W. O. 325
Neurath, H. 350
Neville, J. F., Jr. 329
Newman, E. A. 330
Newton, M. 317, 319, 337
Nichol, E. S. 360
Nicholas, H. J. 345
Nickerson, M. 360, 366
Nieset, R. T. 337
Nightingale, G. 328
Nims, L. F. 316
Noback, C. R. 329
Noble, R. P. 329
Noel, R. H. 358
Nomaguchi, G. M. 360
Noonan, T. R. 336
Northup, D. W. 324, 329
Noshold, D. 370
Novelli, D. 341
Nowinski, W. W. 337
Nulsen, F. E. 331

O

O'Dell, B. L. 374
Oesper, P. 343
Ogden, E. 321
Ogilvie, G. F. 375
Ogle, B. C. 315, 323
Ogur, M. 343
Ohler, E. A. 321
Olcott, H. S. 352
O'Leary, E. 323, 332
Olmsted, F. 332
Olsen, N. S. 321, 326
Olson, R. E. 350
Oppenheimer, E. 361
Oppenheimer, M. J. 329
Orahovats, P. D. 337
Orth, O. S. 361
Orten, J. M. 344
O'Shanny, W. J. 356
Oshlag, A. M. 356
Ostro, M. 354
Otis, A. B. 317, 320
Otto, G. F. 359
Overman, R. R. 323, 370
Owades, P. 347
Owen, S. E. 370

P

Pace, N. 320
Packchanian, A. 378
Packer, H. 377
Page, E. W. 365
Page, I. H. 320, 321, 326, 332, 340
Painter, E. E. 335
Painter, J. 327
Panzarella, F. P. 374
Pappenheimer, A. M., Jr. 378
Parke, H. C. 340
Parkhurst, C. 340
Parkins, W. M. 337
Parkinson, L. R. 373
Parrack, H. O. 335
Parry, T. M. 317
Parsons, H. T. 375
Paschkis, K. E. 320, 333
Patt, H. M. 333, 335
Patterson, E. K. 350
Patterson, H. R. 345
Patterson, J. L., Jr. 334
Patterson, P. 357
Patterson, T. L. 322
Patton, M. F. 360
Paul, W. D. 323, 352
Pauls, F. 340
Peacock, W. C. 328, 375
Pearce, L. 367
Pearlman, W. H. 333, 345
Pearlmutter, M. 363
Pearson, P. B. 375
Peck, H. M. 356
Pecora, L. J. 362
Peiss, C. N. 323
Pelletier, M. H. 357
Penfield, W. 335
Perez, E. J. 376, 377
Perkins, J. F., Jr. 339
Perlman, E. 378
Perlmutter, M. 324
Perlzweig, W. A. 372
Perog, J. 357
Perry, T. M. 320
Pertzoff, V. A. 327
Pessotti, R. L. 364
Peters, J. E. 317
Peters, J. H. 337
Peterson, L. H. 336
Peterson, O. L. 379
Peterson, W. H. 351
Pett, L. B. 375
Pettengill, A. G. 362
Pettinga, F. L. 336
Pfaff, M. L. 351
Pfeiffer, C. C. 355, 356
Phatak, N. M. 355, 366
Philips, F. S. 366
Phillips, J. 355
Phillips, P. H. 373
Pickering, R. W. 366
Pierce, H. B. 375
Pierce, J. 353
Pillemer, L. 377
Pillion, E. L. 319, 339
Pincus, G. 333
Pincus, I. J. 330, 339
Pincus, J. B. 350
Pish, G. 345
Pitts, R. F. 314, 331, 332
Plummer, A. J. 364
Podolsky, B. 327
Podore, C. J. 325, 330
Poel, W. E. 334
Poet, R. 354
Polis, B. D. 342
Pollack, A. A. 317
Polli, J. F. 358
Pomerat, C. M. 327
Pool, J. L. 335
Popper, H. 367, 374
Porter, C. C. 345
Porter, T. 372
Porter, W. B. 343
Porterfield, V. T. 364
Post, R. L. 338
Postel, S. 322
Posternak, J. M. 319, 327
Powell, C. C. 362
Power, M. H. 351
Prado, J. L. 325, 349

Pratt, H. J. 366
Prescott, K. F. 324
Pressman, D. 379
Prestrud, M. C. 333
Price, V. E. 347
Pritchard, J. A. 347
Proffitt, J. E. 377
Prouty, L. R. 328
Pugsley, L. I. 345
Putnam, F. W. 342

Q

Quaife, M. L. 373
Quashnock, J. M. 352
Quick, A. J. 369
Quigley, J. P. 329, 338

R

Rachele, J. R. 348
Racker, E. 343
Radomski, J. L. 362
Ragan, C. 346
Rahn, H. 317, 320, 338
Raidt, H. A. 359
Rake, G. 378
Rakoff, A. E. 320, 333
Rall, D. P. 353
Ralli, E. P. 330, 372
Ralston, H. J. 334
Rampton, S. 345
Ramsaroop, A. 319
Ramsey, R. W. 336
Randak, E. F. 329
Randall, W. C. 323, 332
Ransmeier, R. E. 322
Rapaport, S. I. 319
Rapela, C. 315
Rapport, D. 325, 330
Rapport, M. M. 320
Rasmussen, T. W. 335
Rathbun, J. C. 324
Rawls, K. 344
Rebuck, J. W. 367
Recant, L. 333
Redondo, H. P. 339
Reed, C. I. 323
Reed, E. A. 317
Reed, R. K. 365
Reese, J. D. 379
Regan, M. 347
Reinecke, R. M. 324
Reiner, J. M. 320, 321
Reinhard, J. F. 367
Reinhard, J. J. 321
Reinhard, J. J., Jr. 340
Reiss, R. A. 328, 329
Remington, J. W. 329
Rennick, B. 354
Renshaw, B. 326
Reubi, F. C. 315
Reynolds, M. 324
Reynolds, S. R. M. 318
Rhode, C. M. 337
Rhodes, C. 358
Rice, C. E. 378
Richards, B. 353, 361, 365
Richards, C. H. 335
Richards, D. W., Jr. 329
Richards, R. K. 368
Richardson, A. P. 357, 358
Richardson, G. S. 320
Richardson, J. A. 353
Richardson, L. R. 374, 376
Rigdon, R. H. 370
Riggs, D. S. 361
Riggs, T. R. 372
Riley, R. L. 329, 336
Rinehart, J. F. 344, 369
Ring, G. C. 329
Risley, H. A. 342
Risman, G. C. 336
Rittenberg, D. 344, 347
Robb, J. S. 316
Roberts, E. 351
Roberts, J. T. 331, 364
Roberts, S. 330, 342
Robertson, C. R. 322
Robinson, C. S. 330
Robinson, F. J. 356
Robinson, T. W. 327
Robscheit-Robbins, F. S. 370
Rodbard, S. 323, 338
Roe, J. H. 373
Roemhild, F. 324
Roemmelt, J. C. 331, 332
Rolf, D. 315
Rollins, R. R. 366
Romanoff, L. 333
Romans, F. 349
Roos, A. 329
Rosen, F. 372
Rosenbaum, H. 326
Rosenberg, I. M. 341
Rosenberg, L. 338
Rosenblatt, A. 322
Rosenfeld, S. 321
Rosenow, E. C. 368, 379
Rosenthal, O. 348
Rosenthal, S. M. 363, 378
Roth, F. 365
Roth, G. M. 328
Roth, J. L. A. 323
Roth, L. W. 368
Roth, P. 372
Rothenberg, M. A. 321, 326
Rothstein, A. 343
Rouser, G. 371
Routh, J. I. 323, 352
Roux, R. M. 350
Rubenstein, B. B. 318, 340
Rubin, L. 344
Rubin, M. 373
Rubin, S. R. 376
Rudkin, G. T. 344
Rudolph, G. G. 352
Ruhe, C. H. W. 337
Russel, W. C. 372
Russo, H. F. 356
Ryan, A. H. 323

S

Sachs, E., Jr. 334
Saffran, M. 349
Saidel, L. 322
Salerno, P. R. 357
Salis, H. 347
Salmon, W. D. 372
Salomon, K. 336
Salter, W. T. 318, 325
Saltzstein, H. C. 322
Sampson, F. 323
Samuels, L. T. 345, 352, 376
Samuelsen, C. S. 345
Sanders, F. K. 376
Sandow, A. 334
Sandweiss, D. J. 322
Sano, M. E. 378
Santos-Martinez, J. 355, 356
Sarett, H. P. 372, 375
Sarich, P. 349
Saris, D. 317
Sartorius, O. W. 331, 332
Sauberlich, H. E. 345
Saunders, J. P. 348
Saunders, P. R. 350, 363
Saxey, E. 366
Scantlebury, R. E. 326
Schabel, F. M., Jr. 370
Schachter, R. J. 356
Schade, A. L. 342
Schaffenburg, C. A. 321
Schales, O. 350
Schiess, W. A. 332
Schlamowitz, M. 352
Schmidt, C. F. 322, 355
Schmidt, G. 343
Schmidt, L. H. 359
Schmitt, F. O. 319
Schmitt, O. H. 326
Schneeberg, N. G. 337
Schneider, H. A. 373
Schneider, J. J. 345
Schoepfle, G. M. 319
Schroeder, H. A. 315, 321, 326
Schubert, J. 341
Schubert, M. 359
Schuck, C. 373
Schultz, E. W. 367
Schultz, J. 344, 348, 350, 371
Schwartz, B. S. 356
Schweigert, B. S. 347
Schwimmer, S. 346
Scott, J. C. 317, 339
Scroggin, P. 347
Scudi, J. V. 357, 367
Seager, L. D. 356, 360, 370
Seaman, G. R. 340
Seamans, L. A. 379
Seegers, W. H. 368, 369
Seevers, M. H. 364
Segaloff, A. 369
Seibert, F. B. 351
Seibert, M. V. 352
Seifter, J. 360, 368
Seifter, S. 344, 345
Selle, W. A. 318
Sellers, E. A. 361
Selye, H. 321, 325, 333
Sevag, M. G. 378
Shadle, O. W. 337, 338
Shaffer, C. B. 332, 348, 373
Shaffer, F. 353
Shane, S. J. 363, 364
Shanes, A. M. 319
Shapiro, H. 335, 340
Shapiro, K. L. 329
Sharpe, L. M. 328, 375
Sharples, W. 326
Sharpless, N. E. 362
Shaw, R. S. 327, 339
Shay, H. 322
Sheard, C. 328, 334
Sheldon, W. H. 330
Shelley, W. B. 323, 340
Shellhamer, R. H. 378
Shemin, D. 344
Shepherd, J. 349
Shepperd, I. M. 368
Sherman, M. 352
Sherrod, T. R. 355, 356
Sherwood, R. M. 376
Shettles, L. 318
Shideman, F. E. 358, 366
Shipley, R. E. 336
Shively, R. 374
Shock, N. W. 328
Shorr, E. 315, 321, 329, 330, 340
Shorr, H. M. 330
Shortes, G. 354
Shuler, R. H. 328
Shull, G. M. 351
Shulman, S. 352
Shumacker, H. B., Jr. 331
Sica, A. J. 374
Sichel, F. J. 340
Silber, R. H. 345
Simon, A. 364
Sims, E. S. 347
Sinex, F. M. 346
Sinex, M. 341
Singer, E. 378
Siplet, H. 322
Skeggs, L. T. 351
Slanetz, C. A. 360
Slater, I. H. 324, 325
Sleeth, C. K. 357
Slobodiansky, E. 341
Smadel, J. E. 370, 376
Smetak, E. M. 349
Smith, D. E. 322
Smith, E. 375
Smith, E. L. 350
Smith, H. W. 331, 364
Smith, J. 363, 365
Smith, J. A. 337
Smith, J. K. 373
Smith, J. M. 374, 375
Smith, J. R. 338
Smith, M. I. 359
Smith, M. J. 336
Smith, P. K. 355, 358, 359, 360
Smith, P. N. 368
Smith, R. G. 359
Smith, W. W. 325, 374
Snape, W. J. 331
Snider, R. S. 331
Snyder, F. F. 340
Snyderman, S. E. 349, 376
Sobel, A. 356
Sobel, A. E. 373
Sobel, H. 370
Sodi, E. 361
Sokalchuk, A. 320
Sola, O. M. 334
Solomon, A. K. 346
Solomon, J. D. 344
Somogyi, M. 346
Sonnenschein, R. R. 360
Soodak, M. 341
Spealman, C. R. 317, 338
Spears, J. 369
Speck, J. 350
Spencer, J. N. 317, 320, 335
Sperry, W. M. 343
Spiegel, E. A. 331
Spiegel-Adolf, M. 352
Spiegelman, S. 320, 321
Spivey, M. 375
Sprague, G. F. 362

Springer, B. 346
Sprinson, D. B. 347
Sproul, E. E. 344
Squires, W. 359
Stacy, R. W. 320
Stahl, N. M. 331
Stanbury, J. B. 353
Stannard, J. N. 350
Stare, F. J. 350, 373
Starr, I. 329, 355
Stavraky, G. W. 316, 330, 338, 363
Steers, E. 378
Stefanini, M. 352, 369
Steigmann, F. 358, 367
Stein, W. H. 341
Steiner, A. 371
Stekol, J. A. 348
Stetten, DeW. 351
Stettner, C. E. 320
Stevenson, J. 324
Stickney, J. C. 324, 329
Stimmel, B. F. 345
Stock, I. E. 323
Stoerk, H. C. 369
Stokinger, H. E. 362
Stokstad, E. L. R. 347, 353
Stoll, A. M. 335
Stone, C. A. 353
Stone, D. 333, 333
Stone, F. 334
Strachman, N. 343
Strajman, E. 320
Straus, O. H. 356
Strecker, H. 341
Strobino, L. J. 371
Strob, E. 352
Strominger, J. L. 328
Strong, F. M. 372
Stubbs, J. 318
Stueck, G. H., Jr. 330
Stutzman, J. W. 336
Sullivan, M. X. 342
Summerson, W. H. 347, 348
Sure, B. 349, 374, 376
Surtshin, A. 333
Suskind, M. 320
Sutherland, E. W. 346
Sutherland, V. 364
Suthon, A. M. 350
Sutliff, W. D. 377
Swann, H. G. 320, 320
Swanson, M. A. 343
Swanson, P. 374, 375
Sweat, M. L. 345
Sweeney, H. M. 314
Swendseid, M. E. 347
Swift, M. N. 333, 335
Swigert, V. W. 340
Swinyard, E. A. 361
Sydenstricker, V. P. 344
Szego, C. M. 330
Szmuszkoviez, B. 360

T

Tabor, C. W. 359
Tabor, H. 363
Taliaferro, L. G. 377
Taliaferro, W. H. 377
Talso, P. J. 315
Tarail, R. 366
Tawab, S. A. A. 361
Taylor, H. L. 317
Taylor, J. G. 332
Taylor, R. D. 321, 326
Teasdall, R. D. 316
Teeri, A. E. 372
Tennent, D. M. 356
Terrier, J. C. 332
Terry, R. 370
Teuber, H-L. 333
Thannhauser, S. J. 343
Tharp, C. P. 323, 323
Thayer, S. A. 345, 351
Thienes, C. H. 350, 363
Thomas, B. G. H. 369
Thomas, J. E. 330, 338
Thomas, R. W. 351
Thompson, C. R. 358
Thompson, R. M. 366
Thompson, R. T. 368
Thorn, G. W. 333
Tidwell, H. C. 348
Tieche, H. L. 364
Tillson, E. K. 356
Ting, F. S. 354
Tislow, R. 318, 363
Tobias, J. M. 334, 340
Tocantins, L. M. 368
Toennies, G. 351
Toman, J. E. P. 326, 361
Torda, C. 340, 357
Torpin, R. 338, 364
Totter, J. R. 347
Tracy, J. B., Jr. 337
Trager, W. 369, 371
Travers, J. J. 349
Travis, B. L. 369, 370
Treffers, H. P. 379
Troescher, C. 328
Trossbach, J. 347
Truitt, E. B., Jr. 364
Tucker, E. B. 357
Turman, W. G. 316
Turner, R. A. N. 362
Tuttle, A. H. 323, 337
Tuttle, W. W. 317
Tweedy, W. R. 352
Tyree, E. B. 333, 335

U

Uber, R. 364
Udenfriend, S. 341, 342
Uhl, A. H. 361
Umberger, E. J. 345
Ungewitter, L. 315
Unna, K. 355
Unna, K. R. 357, 365
Uzman, L. L. 342

V

Vandam, L. D. 329
VanderWerff, H. 347
van Harreveld, A. 315
Van Liere, E. J. 324, 324, 329
Van Loo, A. 333
Van Maanen, E. F. 357
Van Middlesworth, L. 337
van Wagenen, G. 346
Varney, R. F. 357
Vars, H. M. 337, 344
Velick, S. F. 346
Vennesland, B. 350
Villee, C. A. 346
Vischer, E. 343
Visscher, F. E. 340
Visscher, M. B. 336
von Oettingen, W. F. 362
Voripaieff, H. 341

W

Wachman, J. 325
Waddell, W. E. 356
Waelsch, H. 347
Wagman, I. H. 334
Wakerlin, G. E. 321, 325, 339
Wakim, K. G. 332
Walcott, W. W. 336
Wald, G. 333
Walden, M. K. 353
Waldron, J. M. 368, 370
Walker, E. 320
Walker, H. A. 357, 358
Walker, R. 325
Walker, S. M. 334
Walkling, A. A. 333
Waller, W. S. 356
Walters, G. R. 323
Walton, R. P. 360
Walton, R. P. 353
Wang, C. C. 330
Wang, S. C. 317
Ward, R. 376
Ware, A. G. 368, 369
Warner, R. C. 371
Warren, J. 376
Warren, M. R. 358
Warren, S. 371
Wastl, H. 321
Watts, D. T. 350
Waugh, D. F. 336
Way, E. L. 359
Wayson, E. E. 331
Webb, J. L. 350, 363
Wei, S. H. 378
Weimar, V. 340
Weiner, H. M. 336
Weinstein, H. R. 337
Weisberg, H. F. 324, 325
Weisiger, J. R. 357
Weiss, C. 371
Weiss, M. S. 341
Weiss, R. 359
Welch, A. D. 347
Welch, K. 335
Wells, I. C. 351
Wells, J. A. 353
Welsh, J. H. 328
Welt, I. D. 374
Wenzel, D. G. 361
Werner, H. W. 358
Wertenberger, G. E. 316
Wertheim, J. M. 337
Wertz, A. W. 373
Wesson, L. G. 331
Wesson, L. G., Jr. 332
West, R. 344
Westerfeld, W. W. 349
Weston, R. E. 326, 328, 333
Weswig, P. H. 375
Wheeler, E. 375
Wheeler, H. E. 325
Wheeler, N. C. 329, 329
White, A. 342, 378
White, H. L. 315, 329
White, J. I. 349
Whitehead, M. H. 356
Whitehead, R. W. 317, 320, 335
Whitehorn, W. V. 316
Whitrock, R. M. 364
Whittier, J. R. 314
Wick, A. N. 340
Wierzuchowski, M. 324
Wiese, H. F. 373
Wight, K. 351
Wiker, A. 331
Wikler, A. 366
Wilber, C. G. 340
Wilcoxon, F. 358
Wiley, J. L. 317, 337
Williams, A. H. 315, 321
Williams, E. 375
Williams, H. H. 343
Williams, J. W. 346
Williams, L. O. 328, 374
Williams, M. 316
Williams, W. L. 366
Willis, A. 320
Wills, G. A. 345
Wilson, D. L. 340
Wilson, H. 345
Wilson, J. W. 373
Wilson, M. 317
Wilson, M. H. 348
Wilson, R. H. 366
Winbury, M. M. 362, 367
Winder, V. 333, 340
Winnick, T. 346
Winslow, J. A., Jr. 329
Winter, C. A. 322
Wintersteiner, O. 351
Winzler, R. J. 351
Wirts, C. W. 331
Witham, A. C. 329
Wittle, E. L. 347
Wolbach, S. B. 369
Wolff, H. G. 340, 357
Wolff, W. A. 353
Wolfson, W. Q. 332
Wollenberger, A. 363
Wong, S. K. 363
Wood, E. H. 314, 317, 320, 321, 332
Wood, H. G. 341
Wood, O. 370
Woodard, G. 361, 362, 367
Woodard, P. H. 324
Woodbury, R. A. 338, 364
Woodford, R. B. 322
Woolley, D. W. 342
Woolsey, C. N. 335
Wright, H. N. 359
Wulff, V. J. 340
Wulzen, R. 340
Wycis, H. T. 331
Wycoff, R. W. G. 378
Wyman, J., Jr. 348
Wynn, W. 317

Y

Yaw, K. E. 379
Yiengst, M. J. 328
Young, N. F. 342
Yu, E. A.-F. 365

Z

Zarrow, M. X. 318
Zepplin, M. 373
Ziegler, J. E. 335
Zorn, E. M. 352
Zucker, M. B. 369
Zweifach, B. W. 321, 330, 340
Zweig, M. 358